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OTHER BUSINESS Other correspondence should be directed to the Secretary, Michigan Entomological Society, 2104 Needham Rd., Ann Arbor, MI 48104.
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Cover photo
Protolithocolletis lathyri Braun

Photo by
Sarah Smith
The importance of reporting distributional records for organisms is not always immediately apparent and such information is frequently overlooked. However, at the basic level, having a more complete understanding of the distribution of a given species can help biologists better understand its natural history; this, in turn, can inform targeted conservation efforts (e.g., Vogt and Cashatt 2007 and references therein). Additionally, when ranges of several members of a genus are known, distributional information might allow insights into the evolutionary history of the group or provide opportunities or directions for testing hypotheses (e.g., Funk and Wagner 1995). Records carelessly dismissed and shoved into the dusty bin of “knowledge for knowledge’s sake”, such as those from an “expected” state, might corroborate single accounts from other states or even provide a breakthrough in understanding the microhabitat of a species where previously unknown (e.g., Skvarla et al. 2014). Similarly, records of species repeatedly taken over time can expose temporal changes, which might subsequently be correlated or explained by external biotic or abiotic forces or changes (e.g., Albrecht 1967). Furthermore, a better understanding of where and whether not a species might occur could contain implications for their use and efficacy as biological control agents (e.g., Martin et al. 2018). Of particular significance are distributional records for predators because they compose a significantly smaller biodensity and therefore are typically encountered with less frequency than herbivorous species, affording less opportunity to observe and study them.

Thus, herein are reported new state records for some predatory true bugs. Paralleling my prior treatment involving new pentatomomorphan records (i.e., Swanson 2018), during the course of my survey of the Nabidae of Michigan (i.e., Swanson 2012), the Phymatinae of Michigan (i.e., Swanson 2013), and the Cimicoidea of Michigan (i.e., Swanson 2016), new extra-Michiganian state records for various cimicomorphan species were discovered among material housed in two major university collections in southern Michigan, as well as my personal collection. Those records form the basis of this treatment.

Materials and Methods

After examining the determined “anthocoroid”, nabid, and phymatine holdings and identifying much of the undetermined U.S. material in two Michigan university collections, the locality data of these specimens was compared with the most recent catalog for Heteroptera found north of Mexico (Henry and Froeschner 1988). Further records were culled after comparing with various post-1988 regional faunistic studies, as well as some pre-1988 references overlooked by the authors of the catalog (see annotations, plus Swanson 2011, 2018). The remaining specimen data are the subject of this treatment.

The identification of all specimens included in this study was rendered or confirmed by me, using Harris (1928) for the nabids, Kormilev (1962) for the phymatines, and my own keys (Swanson, unpublished).
to the United States taxa synthesized from various sources (e.g., Herring 1976, Kelton 1978) for the “anthocoroids”. I have confirmed identifications rendered by H. G. Barber, R. F. Hussey, and D. Punzalan, whereas those “anthocoroid” identifications rendered by T. Lewis and a single phymatine identification rendered by N. Kormiliev have been accepted. All new records pertain to specimens vouchered in one of the collections listed below.

Label data were not copied verbatim, although complete locality information is included. Any additions, changes, or interpretative elements I provide are shown in brackets. Multiple localities are included, where possible, to fortify new records. Distributional or taxonomic notes are offered where deemed necessary or useful.

As mentioned, the authority on which a state having a published record is based belongs to Henry (1988) for the Anthocoridae, although this is supplemented with several other recent distributional accounts (i.e., Lewis et al. 2005). For Cimicidae, the authority belongs to Froeschner (1988a). For the Nabidae, the authority belongs to Henry and Lattin (1988) and Kerzhner and Henry (2008). Reviudiv records are based on Froeschner (1988b, c) and Swanson (2011). An updated distribution within the United States and Canada is included for each species treated herein. Records overlooked or reported subsequently are annotated in the distribution north of Mexico given at the end of each species account, whereas new records herein reported are presented in bold type; thus, this treatment also may be used as a partial compilation of references overlooked in Henry and Froeschner’s (1988) catalog. The abbreviations used for each U.S. state and Canadian province or territory follow the partial compilation of references overlooked or reported subsequently are annotated in the distribution north of Mexico given at the end of each species account, whereas new records herein reported are presented in bold type; thus, this treatment also may be used as a partial compilation of references overlooked in Henry and Froeschner’s (1988) catalog. The abbreviations used for each U.S. state and Canadian province or territory follow the in Henry and Froeschner’s (1988) catalog.

Collections are designated as follows: Daniel R. Swanson, personal collection (DRS); Albert J. Cook Arthropod Research Collection, Michigan State University, East Lansing, Michigan (MSUC); and University of Michigan Museum of Zoology Insect Collection, Ann Arbor, Michigan (UMMZ).

Results

As a result of this investigation, 40 new state records for 25 species in 12 genera in 6 families have been compiled. The following 24 states have new records: CA, CT, GA, IL, KY, LA, ME, MN, MT, NC, NE, NH, NM, NV, OH, PA, RI, SC, VA, VT, WA, WI, WV, WY.

Family ANTHOCORIDAE

Nine new state records are reported for 6 species in 4 genera.


Distribution: USA: CT, FL, IL, MI (Lattin 1999), MO, NC (McPherson and Weber 1981), NY, SC (Ulyshen et al. 2012), TX, WI; Canada: NB, NS, ON, PE, QC, SK (Kelton 1978).


Notes: Carpintero (2002) revived the genus Dasyleptus Reuter, 1872a and transferred this species to it, without any justification. In addition to the lack of explanation, Carpintero (2002) overlooked the fact that the anthocoroid genus was preoccupied by Dasyleptus Peters, 1871 in Mammalia; Chiroptera: Vespertilionidae; Swanson (2019) summarized the situation and supplied a replacement name for Dasyleptus Reuter, 1872a.

Distribution: AR (Chordas et al. 2011), FL, GA, LA, SC, TX, VA.


Notes: This species was previously confused with Orius tristicolor (White, 1879) in the east, but Lewis and Horton (2010) clarified its status; the distribution given below is based on that treatment.

Distribution: USA: AK, CT, CO, IA, ID, KS, MA, ME, MI, MN, MT, ND, NE, NH, NY, OR, PA, SD, UT, VT, WA, WY; Canada: AB, BC, NB, NS, ON, QC, YT.

Distribution: USA: AL, AR (Tugwell et al. 1973, Elkassabany et al. 1996), CA, CO, CT, FL, GA (Kelton 1963), IA, IL, IN, KS, KY (Isenhour and Yeargan 1981), LA, MA, MD, ME (Parshley 1917), MI, MN, MO, MS (Lago and Testa 2000), NC, NE, NH, NJ, NM (Townsend 1892), NY, OH, PA, RI (Parshley 1917), SC, SD (Harris 1937), TN, TX, UT, VA, VT (Parshley 1917), WA (Newcomer 1958), WI, WV (Brown et al. 1988); Canada: AB (Knight 1925, Strickland 1953), BC(?), MB, ON, QC.


Notes: As mentioned above, this species was previously confounded with O. diespeter in the east, but Lewis and Horton (2010) clarified its status; the distribution given below is based on conclusions drawn in that treatment, viz. previous records of O. tristicolor in the east actually referred to O. diespeter and the former species has not been collected east of Nebraska. Maw et al.’s (2000) listing of the Northwest Territories remains unplaced.

Distribution: USA: AK, AZ, CA, CO, ID, NE, NM, NV, OR, TX, UT, WA, WY; Canada: AB, BC, MB, SK, YT.


Notes: Lattin (2007) discussed the distribution of this and several other species of Xylocoris in the United States.

Family CIMICIDAE

One new state record is reported for 1 species in 1 genus.


Distribution: USA: AK (Scudder and Sikes 2014), CA, CO, CT (Parshley 1917), FL (Loy and Regan 1991), IA, IL, MA (Parshley 1917), ME, MI (Swanson 2016), NC, ND (Brown et al. 2009), NE, NH, NY, OR, TX (Sikes and Arnold 1984, Kopachenia et al. 2000), UT; Canada: AB, BC, MB (Maw et al. 2000), NS, ON, QC (Maw et al. 2000).

Family LASIOCHILIDAE

One new state record is reported for 1 species in 1 genus.


Family LYCTOCORIDAE

One new state record is reported for 1 species in 1 genus.


Note: Kelton (1967) questioned the validity of Van Duzee’s (1917a) record from British Columbia.

Distribution: USA: AL, AR (Chordas et al. 2005), CA, FL, GA, IA (Kelton 1967), IL (Blatchley 1926, Kelton 1967), IN, LA (Kelton 1967), MI (Swanson 2016), MO, MS (Kelton 1967), NC, ND (Kelton 1967), NY, OH (Kelton 1967), SC, TX, WI; Canada: BC(?), MB (Kelton 1967, Maw et al. 2000).

Family NABIDAE

Fourteen new state records are reported for 9 species in 3 genera.


Notes: The presence of this introduced Palearctic endemic in various parts of North America has been noted by Barber (1932), Scudder (1961), Lattin (1966), and Wheeler (1976).

Distribution: USA: CA, MI (Swanson 2012), NY, OH, OR, PA, WA; Canada: BC, NS (Wheeler and Hoebeke 2004), ON (Paiero et al. 2003).

**Distribution**: USA: AR (Chordas et al. 2005), CT, IA, IL, IN, MA, MD (Brown and Bahr 2008), MI (Hussey 1922, Swanson 2012), MN, MO, MS, NC, NJ, NY, OH, OK (Drew and Schaefer 1963), PA, RI (Parshley 1917), SD, TN (Lambdin et al. 2003), TX, VA, VT (Parshley 1917), WA (Torre-Bueno 1934), WI, WY: Canada: AB, BC, MB (Larivière 1994), NB, NS, ON, PE, QC, SK.


**Distribution**: USA: AR (Tugwell et al. 1973), CO, CT, DC, DE, FL, IA, ID, IL, IN, KS, KY, MA, MD (Brown and Bahr 2008), ME, MI, MN, MO, MS, NC, NE, NH, NJ, NY, OH, OK (Parshley 1917), NV, NY, OH, OK (Drew and Schaefer 1963), OR (Van Duzee 1917b), PA, RI (Parshley 1917), SC, SD, TN, UT (Knowlton 1935), VA, VT, WI, WV; Canada: AB, BC, MB, NB, NS (Parshley 1923, Maw et al. 2000), ON, PE (Scudder 2008), QC, SK.


**Distribution**: USA: AK (Scudder and Sikes 2014), AR (Tugwell et al. 1973, Chordas et al. 2005), AZ, CA, CO, CT, GA, IA, ID, IL, IN, KS, MA, MD, ME, MI, MN, MO, MS, MT, NC, ND, NE, NH (Parshley 1917), NJ, NM, NY, OH, OK (Smith 1940, Drew and Schaefer 1963), OR, PA, RI (Parshley 1917), SC (Roach 1991), SD, TN, TX, UT, VA, VT.


Distribution: AZ, CO, ID (Harris and Shull 1944), KS, MT, NE, NM, OK, OR, TX, UT, WA.


Distribution: USA: ID (Harris and Shull 1944), NV, OR, UT, WA; Canada: BC.


Notes: The combination of a virtual absence of a lateral notch on the pronotum, a simple rounded connexival outline, simple veins of the hemelytral membrane, and size greater than 6.5 mm will diagnose P. borica among the United States fauna. The elevation of the collection site is probably between 6,500 and 8,000 feet, which corresponds well with previous localities for this species. The specimen reported seems to have been syntopic with Phymata saitieri Kormilev (see below).

Distribution: AZ, CO, ID, NM, UT.


Notes: Given the difficulty associated with phymatine identification (see discussion in Swanson 2013), and the early convolution of Phymata erosa Linnaeus, 1758; Phymata wolffii Stål, 1876; and P. fasciata in partic-
ular, the baseline distribution of this species is gleaned from three revisionary studies: Melin (1931), Evans (1931), and Kormilev (1962). Records prior to these treatments (e.g., Osborn 1894: CA; Townsend 1894: NM; Osborn 1900: OH; Cockrell 1901: NM; Hart and Gleason 1907: IL; Barber 1914: FL; Parshley 1917: CT, MA, RI, VT; Parshley 1922: SD; Blatchley 1926: IN; Hendrickson 1928: IA; Johnson 1930: MA; Knowlton 1931: UT; Torre-Bueno 1931, 1933: FL) are considered suspect and therefore excluded.

Harris' (1937) record for South Dakota is also excluded, as he questionably equated his specimens of "Phymata erosa fasciata" with *P. americana*, the latter likely being the correct identification. Melin (1931) described this subspecies, *Phymata fasciata georgiana* Melin, 1931, from "Carolina", Georgia, and Texas. Evans (1931) greatly extended the range of this subspecies, adding Arkansas, Florida, District of Columbia, Illinois, Kansas, Louisiana, Maryland, Mississippi, Missouri, Nebraska, New Jersey, New Mexico, North Carolina, Ohio, Oklahoma, and Virginia, without including specific locality data. Kormilev (1962) noted examined specimens (as *P. f. fasciata*) from a more southern distribution, mentioning only Arizona, "Carolina", Georgia, Louisiana, Mississippi, and Texas; this is the distribution given by Froeschner (1988b). Despite prior characterization of *P. f. fasciata* as a southern or coastal species, Melin’s (1931) treatment is here vindicated, as I have examined the following corroborative material:


Notes: See notes under the preceding subspecies. The baseline for records of this species comes from the original description (Evans 1931: FL, GA), plus a record added by Kormilev (1962: LA). I have examined the following material corroborating the latter: LOUISIANA: [Bat-Pine Can., Baton Rouge, 24 May 1934, F. E. Lyman, det. D. R. Swanson 2012 [1 ♀] (UMMZ)].

Distribution: FL, GA, LA (Kormilev 1962), SC.


Notes: In the United States, only three species, Phymata saileri Kormilev, 1957; Phymata granulosa texasana Kormilev, 1957; and Phymata rossi Evans, 1931, possess setigerous granules on the body, and the latter species is densely covered with these structures, separating it from the other two more sparsely-covered species. The shallow lateral notch of the pronotum will separate P. saileri from P. g. texasana. Furthermore, the localities seem to corroborate this identity as this western New Mexico locality is nearer to areas where the species is known in Arizona; P. g. texasana, however, is known from extreme southern Texas. The specimens here reported appear to have been syntopic with P. borica (see above).

Distribution: AZ, NM.


Subfamily STENOPODAINAE


Distribution: AL, AR, AZ, CO, FL, GA (Swanson 2011), IL, KS, LA, MD, MO, MS (Swanson 2011), NC, NE, NJ, NM (Swanson 2011), OK, SC (Payne et al. 1968, Swanson 2011), TN, TX, VA.

Acknowledgments

The bulk of the work that went into this study was carried out during my time in the UMMZ, and I am grateful to Mark O’Brien (UMMZ) and Gary Parsons (MSUC) for the privilege of studying the material under their care. I also owe thanks to Tamera Lewis (USDA-ARS, Yakima Agricultural Research Laboratory, Wapato, Washington) and Paul Masonick (University of California, Riverside) for correspondence regarding identified material and state records of some “anthocoroid” and phymatine taxa, respectively. I also greatly appreciate the efforts of two anonymous reviewers, who made me aware of several obscure references and/or overlooked records, thereby significantly improving the utility of this study.

Literature Cited


Lambdin, P. L., J. F. Grant, G. J. Wiggins, and A. Saxton. 2003. Diversity of the true...


Tugwell, P., E. P. Rouse, and R. G. Thompson. 1973. Insects in soybeans and a weed host...


Understanding the potential efficacy of a candidate weed biological control agent has become more prominent in recent years, especially pre-release studies of an agent’s impact on plant performance (e.g., Grevstad et al. 2013, Reddy and Mehelis 2015). Such assessments have been proposed for prioritizing agents for further study or release, in order to enhance the success rate of weed biological control programs (Balciunas and Coombs 2004, McClay and Balciunas 2005).

The seasonal duration and amount of damage that an agent might inflict on a targeted invasive plant will depend in part on the number of agent generations (voltinism) produced in the area of release. The induction of diapause in an insect population is a key component determining the number of generations and seasonality of insect activity. For many insects of temperate zones, photoperiod is a primary cue for diapause induction (Tauber et al. 1986).

Black swallow-wort [Vincetoxicum nigrum (L.) Moench = Cynanchum louiseae Kartesz and Gandhi] and pale swallow-wort [V. rossicum (Kleopow) Barbar. = Cynanchum rossicum (Kleopow) Borhidi] (Apocynaceae-subfamily Asclepiadoideae) are European twining vines introduced into northeastern North America. One candidate agent is the defoliator Abrostola asclepiadis (Denis and Schiffermüller) (Lepidoptera: Noctuidae). The moth reportedly has up to two generations in parts of its native range. We assessed the potential multivoltinism of Russian and French populations of the moth by rearing them under constant and changing photoperiods, ranging from 13:11 to 16:8 hour (L:D). The French population was also reared outdoors under naturally-changing day lengths at a latitude similar to northern New York State. Less than six adult moths emerged, with one exception, for any photoperiod treatment. We expect A. asclepiadis to be univoltine if it were to be released into North America, limiting its potential impact on swallow-worts. It should therefore be given a lower priority for release.

Keywords: Cynanchum rossicum, Cynanchum louiseae, swallow-wort, weed biological control, photoperiod

Abstract

A biological control program is in development for two swallow-wort species (Vincetoxicum, Apocynaceae), European vines introduced into northeastern North America. One candidate agent is the defoliator Abrostola asclepiadis (Denis and Schiffermüller) (Lepidoptera: Noctuidae). The moth reportedly has up to two generations in parts of its native range. We assessed the potential multivoltinism of Russian and French populations of the moth by rearing them under constant and changing photoperiods, ranging from 13:11 to 16:8 hour (L:D). The French population was also reared outdoors under naturally-changing day lengths at a latitude similar to northern New York State. Less than six adult moths emerged, with one exception, for any photoperiod treatment. We expect A. asclepiadis to be univoltine if it were to be released into North America, limiting its potential impact on swallow-worts. It should therefore be given a lower priority for release.
Erebididae) has been released in Canada (2013) and more recently in the U.S. (Weed et al. 2011b, Young and Weed 2014, Milbrath and Biazzo 2016).

One candidate biological control agent is the defoliating moth *Abrostola asclepiadis* (Denis and Schiffermüller) (Lepidoptera: Noctuidae). It is widely distributed in Europe in open field to shaded habitats. In some years and locations it defoliates entire stands of *Vincetoxicum hirundinaria* Medik., its primary host plant (Förare 1995, Leimu and Lehtilä 2006). The moth overwinters as a pupa in leaf litter and has one-two generations per year. Two generations have been reported in some areas of central Europe (Förare 1995) that would have similar latitudes to northern New York and southern Ontario. *Abrostola asclepiadis* appears to be host specific to *Vincetoxicum* spp. (Hazlehurst 2011). However, a single generation appears to be host specific to *V. hirundinaria* (Milbrath et al. 2018, 2019) to growing season in the northeastern U.S. they would typically experience during the growing season in the northeastern U.S. and southeastern Canada. The results were conducted with *A. asclepiadis*.

Our objective was to determine the potential number of generations of different populations of *A. asclepiadis* that could occur in the invasive range of swallow-worts. Populations were exposed to photoperiods they would typically experience during the growing season in the northeastern U.S. and southeastern Canada. The results were to be used in concert with separate impact studies and a population matrix model for swallow-worts (Milbrath et al. 2018, 2019) to predict the efficacy of this candidate agent.

**Materials and Methods**

**Insect cultures.** Eggs of Russian *A. asclepiadis* were collected 29 May–6 June 2013 from *Vincetoxicum* spp. leaves in the Russian North Caucasus, near Kislovodsk (43°56.400' N, 42°30.161' E) and Borgustanskaya (44°02.0436' N, 42°30.161' E). Eggs were transported to the Zoological Institute, Russian Academy of Sciences, St. Petersburg for the first diapause experiment (see Initial diapause test). Diapauing pupae were subsequently shipped to the senior author for additional experiments described below. Eggs of French *A. asclepiadis* were collected 16 June 2014 from *V. hirundinaria* at Glan-dage, France (44°42.033' N, 05°37.000' E). Larvae were reared on *V. hirundinaria* at the European Biological Control Laboratory (EBCL), Montferrier-sur-Lez, France, under a 10:14 hour (L:D) photoperiod to induce diapause and pupae were shipped to the senior author. Voucher specimens have been deposited with the Cornell University Insect Collection, Department of Entomology, Itha-ca, NY (under Lot Number 1263).

Diapausing pupae were overwintered at 5°C and a 10:14 hour (L:D) photoperiod for up to 9 months. Pupae were then transferred to a chamber set at 14:10 hour (L:D) and 25:20°C for adult emergence, mating, and oviposition. Two groups of adult moths were held in 60 × 60 × 60 cm aluminum-mesh cages containing honey-water and potted pale or black swallow-wort plants for oviposition. Leaves with egg masses were collected daily (< 24 hour post-oviposition) to minimize the time at a non-experimental photoperiod.

**Initial diapause test.** Neonate larvae (1st laboratory generation) were placed individually into ventilated 0.25 liter plastic vials and randomly assigned to custom-made programmable chambers set to 20 or 25°C and one of six photoperiod treatments at each temperature (hours, L:D): 13:11, 16:8, 13:11 to 16:8 fast, 16:8 to 13:11 fast, 13:11 to 16:8 slow, or 16:8 to 13:11 slow. A short day of 13 hours represents natural day lengths in the North Caucasus in early-April or early-September, and a long day of 16 hours exceeds the longest summer day length (sunrise to sunset, Fig. 1). For the fast photoperiod change, larvae were transferred to the alternate photoperiod at approximately the third instar or halfway through their larval development (day 11 at 20°C, day 8 at 25°C). For the slow change, day length was increased (or decreased) daily in a stepwise fashion over a 5 day (20°C) or 3 day (25°C) period. Ten larvae were used per treatment combination for each of the two Russian populations for a total of 240 larvae. Larvae were fed excised leaves from *Vincetoxicum* sp. plants that had also been collected from the North Caucasus region. The larvae were checked daily and leaves were replaced. Upon pupation, pupae were maintained under their same experimental rearing conditions for an additional 40 days, which is over twice the expected time for emergence from non-diapausing pupae (L. Milbrath, unpublished data). Live pupae that had not emerged as adults at the end of this period were considered to be in dia-pause. Temperature, photoperiod and source population were treated as a single combined factor and tested against the categorical data of diapause (yes or no) using the G-test with simultaneous test procedures (Sokal and Rohlf 1995).
Constant photoperiods. Based on the most likely conditions to promote non-diapause development from the initial diapause test, we used 20°C for further tests with a Russian (Kislovodsk) or French (Glandage) population. The average monthly temperature in Ithaca, NY from June to August ranges from 18.1–20.4°C (NRCC 2018). The experimental design was a one-way treatment structure in a completely randomized design with five constant photoperiods [14:10, 14.5:9.5, 15:9, 15.5:8.5 or 16:8 hours (L:D)]. A short day of 14 hours represents natural day lengths across New York State and north to Ottawa, ON in late-April or mid-August, and a long day of 16 hours exceeds the longest summer day length (Fig. 1). Populations were tested in different years and consisted of larvae of the 2nd laboratory generation.

Groups of egg masses were randomly assigned to one of the five photoperiod treatments in programmable incubators (model I30BLL, Percival Scientific, Inc., Perry, IA 50220). Egg masses were initially placed into 14.5 × 2 cm Petri dishes lined with moist filter paper. At the black-head stage (c. 5 days), groups of c. 80 fertile eggs were transferred to 27 × 19 × 9.5-cm ventilated plastic boxes for larval rearing with five replicates per photoperiod treatment. Boxes contained a false bottom of plastic mesh to allow frass to collect away from developing larvae and plant leaves. Bouquets of greenhouse-grown pale or black swallow-wort stems in water-filled vials were added. Additional bouquets were added as needed until 3rd instar larvae were present, at which time cut stems of field-collected pale swallow-wort were added daily to maintain an excess of food. Black and pale swallow-wort are both suitable hosts for A. asclepiadis (Weed et al. 2011b). Boxes were cleaned of old stems and frass every few days. Prior to eclosion of 4th instars, larvae were randomly culled to 50–55 larvae per box to minimize cannibalism and food shortages.

Upon pupation of all larvae, boxes were cleaned, cocoons were opened and live pupae were placed back into the boxes on a layer of moist vermiculite. Pupae were maintained under their respective experimental rearing conditions and observed for adult emergence over a 50 day period. Remaining pupae were considered alive but in diapause if there was visible movement of the abdomen when gen-

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Figure 1. Day lengths (sunrise to sunset, USNO 2018) from spring through autumn at select North American and European locations.
tly squeezed with soft forceps. Dead pupae were dissected. If pharate adults were found, the individual was scored as non-diapausing. All other dead pupae were omitted from analyses. The percentage of diapausing *A. asclepiadis* was calculated as: % Diapause = [Number of live pupae/(number of emerged adults + number of live pupae + number of dead pharate adults)] (Miller et al. 2000). Diapausing pupae were subsequently ramped down to 5°C over 4 weeks and overwintered as previously described for use in the next experiment. Percentage diapause data were separately analyzed for the two populations using analysis of variance with a logit transformation (PROC MIXED, SAS Institute 2012). Means were separated using the least-significant difference test (SAS Institute 2012).

**Changing photoperiods.** An experiment was conducted to assess the effect of changing photoperiods on diapause induction for the Russian and French populations (3rd laboratory generation) of *A. asclepiadis*. The experimental design, methods and analysis were identical to those described under **Constant photoperiods** except that two changing and two constant photoperiod treatments were used: 14:10 to 16:8, 16:8 to 14:10, 14:10, and 16:8 hours (L:D). For the changing photoperiod treatments, boxes of larvae were transferred to the alternate photoperiod the day after most larvae in a box molted to 3rd instar.

**Natural photoperiods.** An outdoor observational study was conducted in France at the EBCL (43°41.033’ N, 03°52.500’ E) over a three-year period using insects from the Glandage population. Day lengths at EBCL are comparable to those in northern New York State (Fig. 1). In 2016 and 2017, eggs and 1st instars of *A. asclepiadis* were collected from the field in June and reared to the 2nd instar in Petri dishes in the laboratory at c. 22°C and under natural day lengths (next to a window). Larvae were then placed on potted *V. hirundinaria* plants, 2–4 larvae per pot, covered with a 50 cm tall mesh bag supported by metal rods. Pots were located outdoors in a semi-shaded location where the larvae experienced naturally-changing day lengths. Pots were watered as needed and observed for adult emergence. Pupae were recovered from all pots in August or September and remained outdoors on a covered and potted plant for additional observations. In 2016, plants were infested with a total of 78 2nd instars on 22 June and 6 July. In 2017, plants were infested with 108 2nd instars on 19–26 June.

Pupae from the 2017 experiment were overwintered outdoors at EBCL, and ten adult moths emerged 7–11 May 2018. Moths were maintained outdoors on a potted and covered *V. hirundinaria* plant and eggs were collected, approximately one month earlier than in the field at Glandage. On 17 May, 107 eggs and 72 1st instar larvae were distributed among 20 potted *V. hirundinaria* plants and pots were covered with a mesh bag. Additional foliage was added to the caged plants to allow larvae to complete their development. Pupation was observed to occur beginning 12 June. Pupae were recovered from all pots on 27 July (c. 45 days after first pupation) and transferred to a glasshouse cage containing potted plants for an additional month of observation under natural day lengths.

**Results**

Varying numbers of non-diapausing moths emerged at 20°C, but not 25°C, for both Russian populations (Table 1). The greatest emergence, and therefore generally

<table>
<thead>
<tr>
<th>Temperature (°C)</th>
<th>Photoperiod (hours, L:D)</th>
<th>Kislovodsk</th>
<th>Borgustanskaya</th>
</tr>
</thead>
<tbody>
<tr>
<td>20</td>
<td>13:11</td>
<td>60 (10) ab</td>
<td>100 (10) a</td>
</tr>
<tr>
<td></td>
<td>16:8</td>
<td>100 (10) a</td>
<td>100 (10) a</td>
</tr>
<tr>
<td></td>
<td>13:11 to 16:8 fast</td>
<td>90 (10) ab</td>
<td>90 (10) a</td>
</tr>
<tr>
<td></td>
<td>16:8 to 13:11 fast</td>
<td>20 (10) b</td>
<td>56 (9) ab</td>
</tr>
<tr>
<td></td>
<td>13:11 to 16:8 slow</td>
<td>100 (9) ab</td>
<td>100 (9) a</td>
</tr>
<tr>
<td></td>
<td>16:8 to 13:11 slow</td>
<td>100 (10) a</td>
<td>100 (10) a</td>
</tr>
<tr>
<td>25</td>
<td>13:11</td>
<td>100 (10) a</td>
<td>100 (10) a</td>
</tr>
<tr>
<td></td>
<td>16:8</td>
<td>100 (10) a</td>
<td>100 (9) ab</td>
</tr>
<tr>
<td></td>
<td>13:11 to 16:8 fast</td>
<td>100 (9) ab</td>
<td>100 (10) a</td>
</tr>
<tr>
<td></td>
<td>16:8 to 13:11 fast</td>
<td>100 (10) a</td>
<td>100 (10) a</td>
</tr>
<tr>
<td></td>
<td>13:11 to 16:8 slow</td>
<td>100 (9) ab</td>
<td>100 (10) a</td>
</tr>
<tr>
<td></td>
<td>16:8 to 13:11 slow</td>
<td>100 (10) a</td>
<td>100 (10) a</td>
</tr>
</tbody>
</table>

* Values followed by the same letter are not significantly different (G-test with simultaneous test procedure performed on counts, P > 0.05).
the least percentage diapause, occurred with the Kislovodsk population under the “16:8 to 13:11 fast” treatment. Some appreciable emergence also occurred for this same treatment for the Borgustanskaya population and under a short day (13:11) for the Kislovodsk population (Table 1).

In subsequent tests with daylengths ranging from 14 to 16 hours and much larger numbers of larvae, 0–5 adults of the French or Russian populations emerged from the various constant or changing photoperiod treatments. Percentage diapause was therefore 98–100% across the different tests (Table 2). In outdoor tests in France, one adult moth emerged from 18 surviving pupae in 2016 (94% diapause), no adults emerged from 38 pupae in 2017 (100% diapause), and no adults emerged from 95 pupae in 2018 (100% diapause).

Discussion

Both the Russian (Kislovodsk) and French (Glandage) populations of A. asclepiadis appear to be univoltine under photoperiods they will experience during the spring and summer in the northeastern USA and southeastern Canada (Fig. 1). The fact that the two geographically separated populations in our study showed a similar photoperiodic response suggests that other populations that may be considered for release (e.g., Ukraine, Hazlehurst 2011) would also likely be univoltine, at least under the experimental conditions we provided. In Europe, A. asclepiadis appears to be a primarily univoltine species although bivoltine populations have been reported (Förare 1995, Weed et al. 2011a). Voltinism among other temperate noctuid species in the same subfamily (Plusiinae) as A. asclepiadis is variable (Saulich et al. 2017). Some species are strictly univoltine with an obligate diapause, e.g., Charanyca trigrassimica (Hufnagel) (Sokolova 2007). Other species have a facultative diapause and variable numbers of generations depending on the latitude or when offspring are produced in the spring relative to current photoperiods, e.g., the congener A. triplasia (L.) (Saulich et al. 2015).

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In this study we did not determine the critical day length for diapause induction in A. asclepiadis, i.e., the day length that results in 50% of the individuals in a population entering diapause. However, a small pilot study in which larvae of the Kislovodsk population were reared at 18:6 hour (L:D) resulted in only 60% diapause (n=10, L. Milbrath, unpublished data). This result and the significant emergence of adult moths for a few treatments in the first diapause test (Table 1) suggest that the Russian populations of A. asclepiadis have a facultative, and not an obligate, diapause (Sokolova 2007). Nevertheless, the experimental conditions observed to promote non-diapause development (>16 hours day length, a rapid 3 hour decrease in day length) are ecologically unrealistic. No such conditions naturally occur in either the area of moth collection or intended introduced range (Fig. 1). The French population may also have a facultative diapause, but the conditions which might promote a second generation are unknown. Rearing the French population outdoors at a typical time (2017 test) or even one month earlier than normal (2018 test) did not result in a (partial) second generation.

Table 2. Mean (± SD) percentage diapause (total live insects assessed) for two source populations of Abrostola asclepiadis reared under different constant and changing photoperiod regimes, 20ºC 1.

<table>
<thead>
<tr>
<th>Photoperiod (hours, L:D)</th>
<th>Glandage, France</th>
<th>Kislovodsk, Russia</th>
</tr>
</thead>
<tbody>
<tr>
<td>Constant</td>
<td></td>
<td></td>
</tr>
<tr>
<td>14:10</td>
<td>99.3 ± 1.5 (171)</td>
<td>100 ± 0 (219)</td>
</tr>
<tr>
<td>14:5:9.5</td>
<td>99.5 ± 1.0 (198)</td>
<td>100 ± 0 (196)</td>
</tr>
<tr>
<td>15:9</td>
<td>100 ± 0 (189) a</td>
<td>100 ± 0 (197)</td>
</tr>
<tr>
<td>15:5:8.5</td>
<td>99.5 ± 1.1 (199)</td>
<td>100 ± 0 (181)</td>
</tr>
<tr>
<td>16:8</td>
<td>100 ± 0 (196) a</td>
<td>100 ± 0 (140)</td>
</tr>
<tr>
<td>F4, 20 = 0.50; P = 0.735</td>
<td></td>
<td>No analysis possible</td>
</tr>
<tr>
<td>Changing</td>
<td></td>
<td></td>
</tr>
<tr>
<td>14:10 to 16:8</td>
<td>99.1 ± 2.0 (196)</td>
<td>97.8 ± 1.3 (192) b</td>
</tr>
<tr>
<td>16:8 to 14:10</td>
<td>99.5 ± 1.1 (171)</td>
<td>100 ± 0 (203) a</td>
</tr>
<tr>
<td>14:10 control</td>
<td>100 ± 0 (201) a</td>
<td>100 ± 0 (218) a</td>
</tr>
<tr>
<td>16:8 control</td>
<td>97.6 ± 3.0 (183)</td>
<td>99.6 ± 1.0 (223) a</td>
</tr>
<tr>
<td>F3, 16 = 1.80; P = 0.188</td>
<td></td>
<td>F3, 16 = 7.57; P = 0.002</td>
</tr>
</tbody>
</table>

1 Within each test and population, individual means followed by the same letter are not significantly different (F-protected LSD test with logit-transformed data, P > 0.05; n=5).
It is unknown if a change in critical day length for diapause induction would occur in *A. asclepiadis* should it be released into North America, allowing for two generations. The critical day length for the biological control agent *Diorhabda carinulata* (Desbrochers) (Coleoptera: Chrysomelidae) was documented to decrease following release, allowing it to extend its range to more southern latitudes (Bean et al. 2012). It is also unknown if sufficient degree-days are available to complete development to the (overwintering) pupal stage should a second generation of *A. asclepiadis* occur in the future. Two generations of *A. asclepiadis* are reported from central Europe and an occasional partial second generation in Sweden is thought to occur, perhaps due to differing seasonal temperatures or critical day lengths (Förare 1995).

The potential impact of *A. asclepiadis* appears to be limited with a univoltine life cycle. Available defoliation impact data of *A. asclepiadis* (Milbrath et al. 2019) combined with swallow-wort population models (Milbrath et al. 2018) suggest that a single complete defoliation will cause population declines in only a limited number of slow-growing forest and field infestations of pale and black swallow-wort. This contrasts with the released agent *H. opulenta* that will likely have two generations per summer (Weed and Casagrande 2010); it is projected to control more populations of swallow-wort due to the greater amount of damage it can potentially inflict. Agents that have multiple generations, and therefore can provide season-long damage of targeted weeds, are preferred because they limit the ability of plants to recover. This is particularly true for perennial plants (Syrett 1983, Hosking et al. 1994, Winston et al. 2014). As also noted by Milbrath et al. (2019), direct competition between *A. asclepiadis* and *H. opulenta* is likely and should be avoided. For these reasons, *A. asclepiadis* should be considered a low priority agent for release.

**Acknowledgments**

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Saulich, A.Kh., I.V. Sokolova, and D.L. Musolin. 2015. Seasonal development of the dark spectacle Abrostola triplasia (L.) (Lep-
Lessons Learned: Rearing the Crown-Boring Weevil, *Ceutorhynchus scrobicollis* (Coleoptera: Curculionidae), in Containment for Biological Control of Garlic Mustard (*Alliaria petiolata*)

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Abstract

In this paper, we describe lessons learned and protocols developed after a decade of rearing *Ceutorhynchus scrobicollis* Nerenschéimer and Wagner (Coleoptera: Curculionidae) in a Biosafety Level 2 containment facility. We have developed these protocols in anticipation of approval to release *C. scrobicollis* in North America for the biocontrol of garlic mustard. The rearing protocol tried to minimize the potential of attack by the adult parasitoid, *Perilitus consuetor* Nees (Hymenoptera: Braconidae), which may be present in field collected *C. scrobicollis* from Europe to prevent inadvertent introduction of parasitoids into North America.

All *C. scrobicollis* used for our quarantine rearing were field collected near Berlin, Germany. We have successfully reared *C. scrobicollis* on caged garlic mustard plants in a growth chamber by alternating temperatures and photoperiods to simulate those in its native range. In Germany, *C. scrobicollis* produces one generation per year and F1 adults emerge in late May. In containment, a new generation of adults emerged an average of 108 days after adults were placed on plants. We found the optimal time spent to collect F1 adults was four weeks after the appearance of the first F1 adult, with 95% of potential adults collected. Simulating a three-month summer aestivation period, followed by a week of fall, and three weeks of winter conditions resulted in optimum levels of oviposition in F1 females. Larvae first hatched 8- to-10 days after adults were placed on plants at 15/14 C day/night temperatures with a 9.5 hour photoperiod. We therefore recommend that *C. scrobicollis* adults are removed from garlic mustard rosettes after 8 days. This will maximize the period of female oviposition while minimizing the time when larvae are available for attack from *P. consuetor*.

Keywords: *Ceutorhynchus scrobicollis*, *Alliaria petiolata*, garlic mustard, biological control of weeds

Garlic mustard (*Alliaria petiolate* (Bieb.) Cavara & Grande) is a biennial plant in the family Brassicaceae, native to Europe, where it has historically been valued for its medicinal and herbal properties (Grieve 1971). It was first recorded in North America in 1868 (Nuzzo 1993). Since its introduction, garlic mustard has spread to the Northeast, Midwest, and the Pacific Northwest, and is scattered throughout the remaining western United States (Nuzzo 1993). Garlic mustard is now recorded in 37 states in the U.S. and 5 Canadian provinces (USDA NRCS 2019) and has the potential for wider distribution (Welk et al. 2002). The plant is listed as a noxious weed in eight states in the U.S. (USDA NRCS 2019).

With the capacity for abundant seed production, garlic mustard can rapidly colonize mesic forests to produce dense stands (Meekins and McCarthy 2002) (Fig. 1) and become more competitive than other woody understory species (Meekins and McCarthy 1999) which may reduce native plant diversity (Stinson et al. 2007). The invasion of garlic mustard into native plant communities can disrupt the mutual associations between native tree seedlings and arbuscular mycorrhizal or ectomycorrhizal fungi (Roberts and Anderson 2001, Stinson et al. 2006, Wolfe et al. 2008, Anderson et al. 2010) that are critical for tree growth and survival and can disrupt native legume-rhizobia mutualism (Portales-Reyes et al. 2015).
Garlic mustard thrives in the forest understory and grows among desirable native plants. Current garlic management strategies include eradicating new populations by the labor intensive and expensive methods of hand pulling or cutting. Application of herbicides can result in non-target injury to native plant species (Becker et al. 2013).

Currently, three *Ceutorhynchus* Germar (Curculionidae) species are under investigation as potential biological control...
agents. Extensive host specificity testing with the crown-mining weevil, *Ceutorhynchus scrobicollis* Nerenschheimer and Wagner has been completed at CABI in Delémont, Switzerland and at a Biosecurity Level 2 (BSL 2) High Containment Facility at the University of Minnesota, St. Paul (Gerber et al. 2009). Results reported in Gerber et al. (2009) and our additional work since (unpublished data) confirm that *C. scrobicollis* is a highly specific herbivore. The USDA APHIS-PPQ Technical Advisory Group for Biocontrol of Weeds (TAG) has reviewed our host range test results and in February 2017 recommended *C. scrobicollis* for release in the U.S. The next steps in the approval process are to ensure compliance with the Endangered Species Act through U. S. Fish and Wildlife Service (USFWS) and the National Environmental Policy Act (NEPA) through the Animal Plant Inspection Service (APHIS) as well as tribal compliance. In Canada, a similar petition was approved in June 2018 and *C. scrobicollis* was subsequently released in the field.

*Ceutorhynchus scrobicollis* is native to central and eastern Europe and its range extends to the eastern Caucasus region and Ukraine (Colonnelli 2004, Rauth et al. 2011). In Europe, *C. scrobicollis* is only recorded from garlic mustard and field attack rates can reach 100%. Plant height, winter rosette survival and seed production are reduced as a result of *C. scrobicollis* attack (Gerber et al. 2007a, 2007b).

In the field, *C. scrobicollis* produces one generation per year. Oviposition begins in early fall, continues throughout the winter and ends in early spring (Gerber et al. 2009) (Fig. 2). Oviposition ceases if the mean daily temperature drops below –5°C (Gerber et al. 2009). Females lay the majority of eggs directly under the leaf epidermis and in leaf petioles of rosettes (Fig. 3), with a smaller number of eggs laid under the epidermis of the root/crown interface (Gerber et al. 2009).

![Figure 2. Life cycle of *Ceutorhynchus scrobicollis*. (Redrawn figure, original figure courtesy of Esther Gerber, formerly of CABI Switzerland). Colored bars indicate when life stages are present or active.](https://scholar.valpo.edu/tgle/vol52/iss2/1)

![Figure 3. *Ceutorhynchus scrobicollis* egg in margin of garlic mustard leaf. Eggs are deposited directly under the leaf epidermis. (Photo: Elizabeth Katovich, University of Minnesota).](https://scholar.valpo.edu/tgle/vol52/iss2/1)
In the field, adult feeding is insignificant. Larvae progress through three instars, which can be distinguished by the diameter of the head-capsule. In Switzerland, first instar larvae are initially found in late September and by early November, third instar larvae are present. All three instars overwinter in roots and crowns of garlic mustard rosettes and the majority of damage to crowns is caused by larval tunneling. By late April, larvae exit garlic mustard roots and crowns to pupate in the soil. New adults (Fig. 4) emerge from early May to mid-June, feed briefly on garlic mustard leaves, then aestivate for the remainder of the summer (Gerber et al. 2009). Feeding and larval tunneling by *C. scrobicollis* can increase mortality of overwintering rosettes. Alternatively, primary shoots of rosettes can be killed, releasing crown buds from apical dominance. This may result in growth of secondary shoots that are thinner, shorter and produce fewer seeds (Gerber et al. 2007a, 2007b). In captivity, adults may live for one to two years, and up to three oviposition periods have been recorded (Gerber et al. 2009).

*Perilitus consuetor* Nees (Hymenoptera, Braconidae) has been identified as an endoparasitoid of *C. scrobicollis* adults (Haeselbarth, unpublished), which is its only host. Parasitism rates of up to 20% have been observed in field collected *C. scrobicollis* adults (Gerber et al. 2009). In Switzerland, *P. consuetor* pupae leave their hosts in May and adult parasitoids emerge by late May to mid-June. It is thought that *P. consuetor* adults attack *C. scrobicollis* in the spring or fall (Gerber et al. 2009). We wanted to prevent the inadvertent introduction of parasitoids into North America so while developing a rearing protocol we tried to minimize the potential of attack by the adult parasitoid, *P. consuetor*, which may be present in field collected *C. scrobicollis* from Europe.

Problems encountered with rearing biocontrol insects can become a major obstacle for a weed biological control program (De Clerck-Floate et al. 2008). In this paper we describe garlic mustard propagation methods and *C. scrobicollis* rearing protocols developed in our BSL 2 facility at the University of Minnesota in anticipation of permission to release *C. scrobicollis*. In addition, we conducted experiments to characterize *C. scrobicollis* development in containment to develop the most efficient and consistently reliable methods to rear *C. scrobicollis* from garlic mustard plants, informed and refined over a decade of experience. A method was also developed to screen for the endoparasitoid, *P. consuetor*, from new generations of *C. scrobicollis* to ensure that any potential parasitoids can be eliminated prior to field release in North America.

Figure 4. *Ceutorhynchus scrobicollis* adult. Actual length, 3- to 4-mm. (Photo: Harriet Hinz and Esther Gerber, CABI Switzerland, Bugwood.org).
Materials, Methods, and Results

Garlic mustard propagation. Garlic mustard plants were propagated from seed to support *C. scrobicollis* rearing efforts. Whenever possible, garlic mustard seeds were stratified and plants propagated outdoors, so that they were phenologically similar to field grown plants. However, since *C. scrobicollis* was reared in containment, it was necessary to have a continuous supply of seedlings and plants year round, so seeds were also stratified at 4°C in a cold room and plants propagated in a greenhouse.

All seeds for garlic mustard propagation were collected from Silver View Park, in Mounds View, MN (Lat: 45° 06’ 22” N, Long: 093° 13’ 00” W). Seeds were cleaned and stored at 4°C. Garlic mustard seeds require cold stratification to break dormancy (Baskin and Baskin 1992). Field stratification consisted of planting seeds in plug trays filled with a standard commercial potting mix (LC8; 70–80% Canadian sphagnum peat moss, 20–25% perlite, 5–10% vermiculite; Sungro Horticulture, Agawam, MA). Trays were placed outside in November in St. Paul, MN and lightly mulched with straw to overwinter. Mulch was removed in early spring (April in Minnesota) when seedlings emerged.

Seeds were stratified in the lab by adding moistened sand to a 90 mm diameter × 15 mm deep plastic petri dish, adding a layer of seeds, then covering the seeds with additional moist sand. Petri dishes were sealed and placed in a refrigerator at 4°C (Baskin and Baskin 1992). After four months, seeds were removed and planted in a plug tray filled with the standard Sungro potting mix described previously. These methods ensured a continuous, year-round supply of seedlings.

Seedlings from both field and lab stratification methods were transplanted into 3.8 l pots containing a commercial rice hull growing mix (BM7; 35% bark; 20% rice hulls; 45% Berger Peat Moss mix, Saint-Modeste, Quebec, Canada). This soil mix was selected because it was purported to provide excellent drainage. We used this soil mix until it was discontinued by the manufacturer. We now use a 30% loam; 30% course sand; 40% peat moss mix provided by our greenhouse services.

Depending on the season, plants were grown outside in a shaded area, or in a greenhouse with a 16 h photoperiod and 21/18°C day/night temperatures. Plants were fertilized with a slow-release fertilizer containing macro- and micro-nutrients (Osmocote Plus, 15-9-12 plus micronutrients, Scotts Company, Marysville, OH) at the recommended rate. Plants were watered only as needed and care was taken not to overwater plants as this promoted root and foliar diseases. Rosettes were a minimum of three months old when they were used for *C. scrobicollis* rearing.

Aphids were a major problem encountered when propagating garlic mustard in the greenhouse. Secondary pests included the diamondback moth (*Plutella xylostella* L.). We avoided applying insecticides for pest control because they could adversely affect *C. scrobicollis*. To reduce insect problems, multiple garlic mustard potted plants were reared in the greenhouse inside large screened cages 2.4 × 0.9 × 0.9 m, length × width × height, respectively (Fig. 5). These cages consisted of frames built from PVC pipe designed to fit inside a greenhouse bench. “No-see-um” polyester netting was used to construct the screen cages that were placed over the PVC frames. The edges of the cages were secured by folding the netting underneath the frames. Ladybugs (*Hippodamia convergens* Guérin-Méneville) were purchased and placed into the screen cages for aphid control.

Field collection and rearing of *C. scrobicollis* in a containment facility. In our BSL 2 containment facility in Minnesota, all *C. scrobicollis* were reared in growth chambers (Model GR-48, Environmental Growth Chambers, Chagrin Falls, OH, 44022; Model ES, Conviron, Pembina, ND, 58271). Incandescent and fluorescent lighting provided an average light intensity of 250 µmol m$^{-2}$ s$^{-1}$, similar to the shaded conditions in the outdoor propagating area. *Ceutorhynchus scrobicollis* were reared on individually potted garlic mustard plants covered with a screen cage made of “no-see-um” polyester netting. The netting was placed over wire loops stuck inside the pot at right angles, secured with elastic, and extended approximately 45 cm above the pot surface (Gerber et al. 2009). Plants were placed on plastic saucers and sub-irrigated.

*Ceutorhynchus scrobicollis* adults were field collected during their oviposition period in the fall, (usually October) in the vicinity of Berlin, Germany (52° 25’ 8.6592”N, 13° 11’ 13.4952”E). Adults were shipped to our BSL2 containment facility at the University of Minnesota. Shipment sizes varied, but always exceeded the 27 adult minimum, the number of individuals required to capture 99% of the genetic diversity at the Berlin collection site (Rauth et al. 2011). Collecting sufficient quantities to capture the entire genetic diversity of adults at a site increases the robustness of subsequent host range tests (Rauth et al. 2011). After arrival in our containment facility, adults were marked with different colored paint pens to easily dif-
Figure 5. Screen cage used in greenhouse to reduce invertebrate pest problems when rearing garlic mustard plants. (Photo: Elizabeth Katovich, University of Minnesota).

Table 1. Conditions in growth chambers to simulate seasons for the rearing of Ceutorhynchus scrobicollis in a Biosafety Level 2 High Containment Facility, University of Minnesota, St. Paul. 2003-2017.

<table>
<thead>
<tr>
<th>Simulated season</th>
<th>Temperaturea (C)</th>
<th>Photoperiodb (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Day</td>
<td>Night</td>
</tr>
<tr>
<td>Fall</td>
<td>18</td>
<td>15</td>
</tr>
<tr>
<td>Winter</td>
<td>15</td>
<td>14</td>
</tr>
<tr>
<td>Spring</td>
<td>18</td>
<td>15</td>
</tr>
<tr>
<td>Summer</td>
<td>21</td>
<td>20</td>
</tr>
</tbody>
</table>

a Relative humidity was maintained at 60–70%.
b Light and dark photoperiod total of 24 hours.
every two to three weeks. The prolonged oviposition period, in combination with the addition and removal of adult pairs to new garlic mustard plants, allowed us to increase the number of plants with eggs for rearing F1 adult progeny. This protocol allowed us to maximize the number of reared F1 adults, despite relatively low adult emergence in growth chamber conditions.

For rearing, typically not more than three pairs of adults were placed on each caged garlic mustard plant. Gerber et al. (2007b) reported that more than four mating pairs reduced number of F1 progeny, as a result of intraspecific competition among larvae. All plants were numbered, and the dates and number of adults added to and removed from each plant were recorded. Adults were removed from plants by sifting through the top layer of soil and manually removing with forceps.

After oviposition, potted garlic mustard plants were maintained in winter conditions until first adult emergence was noted. During that time, rosette top-growth often died back after *C. scrobicollis* larvae mined roots and crowns (Fig. 6), frequently resulting in new lateral shoots arising from crown buds (Fig. 7). Since adults emerging from pupae in the soil litter can be very difficult to find, the first appearance of windowpane feeding on newly expanding garlic mustard leaves was our indicator of when F1 adults (adult progeny of a cohort) had emerged and could be collected. This characteristic “windowpane” was created in the leaf after adults grazed on the epidermis and mesophyll on one leaf surface while the other side remained intact (O’Day et al. 1998) (Fig. 8).

To continue their development, newly emerged F1 adults were removed and placed on new garlic mustard plants, approximately 6 to 8 adults per pot, for a minimum of two weeks in “spring” conditions (Table 1). F1 adults were then placed into three months of “summer” conditions (Table 1) for aestivation followed by one week of “fall” and three weeks of “winter” to facilitate the onset of oviposition. The number and date of F1 adults collected from each plant was...
Figure 7. Regrowth from crown buds of a garlic mustard rosette after *C. scrobicollis* attack. (Photo: Elizabeth Katovich, University of Minnesota).

Figure 8. Adult *Ceutorhynchus scrobicollis* windowpane leaf feeding on caged garlic mustard plant. (Photo: Ghislaine Cortat, CABI Switzerland).
recorded. *Ceutorhynchus scrobicollis* live for up to three years (Gerber et al 2009), so if all adults are not found and removed in each cycle, different age structures will evolve on individual plants.

**Adult *C. scrobicollis* collection and recovery with emergence traps.** Finding newly emerging adults is tedious and time consuming, so we designed emergence traps to collect and recover these F1 adults. The emergence traps were modified from a design used by Skinner et al. (2004) and were created from polypropylene funnels (Nalgene™, Thermo Fisher Scientific, Inc., 168 Third Ave., Waltham, MA 02451) that measured 150 mm × 137 mm × 27 mm, top diameter of spout × total height × stem diameter, respectively. The exterior of the funnel was spray painted black leaving the stem unpainted. The inside of the funnel was scored with a narrow tip on a Dremel® rotary tool (Robert Bosch Tool Co., Mount Prospect, IL. 60056) so that adult weevils could crawl up into the funnel. The traps were sized so that when inverted, the mouth of the funnel fit snugly inside the pot resting on the soil surface covering the opening of each pot containing a garlic mustard plant (Fig. 9).

To attract adults emerging from the soil to the emergence trap, a freshly harvested garlic mustard leaf was placed inside a syringe container (2.5 cm dia × 10.2 cm long polypropylene syringe container for 12 cc Monoject® syringe, without a needle, luer-lock tip, Monoject®, Sherwood Medical Co. St. Louis, MO 63103) and placed over the stem of the funnel. A freshly harvested garlic mustard leaf was kept in place with a 5 mm length cut from a tube of foam pipe insulation for a 1.27 cm diameter pipe, the circumference of the cut piece shortened to a diameter that fit snugly in the syringe container. This arrangement prevented leaves from falling out of the syringe container. Before installing the emergence trap, any remaining green garlic mustard rosette leaves or stems left after larval development were removed from the pot. Since we are working in a quarantine facility, the entire trap was covered with “No-see-um” polyester netting screen cage and secured with elastic around the top of the pot (not shown in Fig. 9). Pots were returned to the growth chamber in winter conditions. During the adult emergence period, traps were checked every two to three days and fresh garlic mustard leaves were placed in the collecting vial, at which time collected adults were removed from the trap and placed onto new garlic mustard plants to feed.

On average, 78% of F1 adults were recovered from traps over the period of adult emergence. This percentage is similar to the average weevil recovery rate from screened plants at CABI in Switzerland. Although not all F1 adults were collected in the emergence traps, it is clearly a more efficient collection method than the alternative of hand sifting through the soil and leaf litter of each individual plant.

**Generation time and number of adults produced in two successive cohorts of *C. scrobicollis*.** Two successive *C. scrobicollis* cohorts were followed to determine the number of days to emergence of F1 adults, total number of adult progeny produced per cohort, as well as average number of adults produced per plant. Cohort 1 adults were field collected in Germany and received in our containment facility and Cohort 2 was the F1 progeny of Cohort 1. Cohorts 1 and 2 F1 adult progeny were collected from 78 and 115 potted garlic mustard plants, respectively, after the repeated transfer of mating pairs.
during *C. scrobicollis* oviposition in our BSL2 facility (as described previously).

For Cohort 1, F1 adult progeny emerged after an average of 106 days (n = 78, SE = 1.9) from the time parents were placed on plants, ranging from 77 to 144 days (Table 2). An average of 4.4, F1 adults emerged from each plant (n = 78 plants, SE = 0.4) with a range of 1 to 16 adults per plant. We reared 347 F1 progeny. For Cohort 2, adults emerged after an average of 110 days (n = 115 plants, SE = 1.5) ranging from 75 to 162 days (Table 2). An average of 4.7 adults emerged per plant (n = 115, SE = 0.5), with a range of 1 to 31 adults per plant. We reared 539 F2 progeny. Generation time from parents to F1 adults averaged 108 days between the two cohorts (Fig. 10). In field reared F1 adults in Delémont, Switzerland, generation time varies, depending on whether eggs are laid in fall, winter or early spring. F1 adults emerge in late May to early June.

Adult emergence in this study was lower than the average of 6.2 adults per plant, recorded for our host range tests conducted earlier under similar conditions, but were highly variable (Katovich, unpublished). The number of *C. scrobicollis* adults recovered from each plant is also lower than reported by Gerber et al. (2009), with an average of 9.2 adults emerging when 2 to 3 females were placed on caged plants for 2- to 4-weeks. Differences in adult emergence between common garden experiments (Gerber et al. 2009) and our results in containment could be the result of less than optimal conditions in growth chambers compared to field settings. Regardless, the repeated transfer method used during oviposition ensured we obtained an increase in *C. scrobicollis* through the multiplier effect of numerous plants exposed to ovipositing females.

**Minimizing the number of weeks required to collect *C. scrobicollis* F1 adults.** F1 adults emerge over a period of time, so collecting adults from caged plants becomes a laborious, time consuming process. To determine the optimum number of weeks required to collect the majority of F1 adults from caged plants, F1 progeny from a single cohort were collected over the course of their entire emergence period. In this cohort of 71 plants, all parent *C. scrobicollis* had been removed within two to three weeks after initial placement on each plant, which ensured that eggs were of similar age. After F1 progeny were first found, funnel traps were placed on plants and checked every two to three days. Emerging F1 adults were removed and numbers recorded. Traps were checked until adults were no longer found.
for two successive collections for each pot. At the end of the collection period, the base of each plant, as well as the surface soil and litter layer, were searched for F1 adults that were added to the number of F1’s collected from funnel traps.

All F1 adults had emerged from pots within six weeks after progeny were first recorded and by week four, 95% of adults had emerged (Table 3). Based on these results, we recommend checking plants for four weeks after the first F1 adult is noted. After five or six weeks post emergence, searching for the remaining 5% of F1 adults is not an optimum use of time and labor. Although we determined the length of F1 emergence in only one cohort of *C. scrobicollis*, these time periods provide guidelines for future rearing efforts.

### Results of the following experiments helped optimize *C. scrobicollis* development and rearing in containment.

#### Experiment 1. Soil medium providing optimum emergence of *C. scrobicollis*

We encountered problems with F1 adult emergence in our growth chambers when using a commercial rice hull soil mix (described previously). Third instar larvae of *C. scrobicollis* exit garlic mustard crowns and use the surrounding soil to build soil pupal chambers. Since the crowns had extensive larval tunneling, we hypothesized that few larvae or pupae were surviving in the soil to emerge as adults. A study was designed to determine the best soil mix to ensure pupa survival and maximize adult emergence. Two treatments tested were 1) a standard rice hull potting mix used to propagate garlic mustard (described previously) and 2) addition of approximately 4 cm of a standard greenhouse soil mix (silt loam: sand: manure: peat, 1:1:1:1, v/v/v/v) covering the soil of the potted garlic mustard plant. Each treatment was replicated 11 times and randomly assigned to a single caged plant as a replicate. Three pairs of marked *C. scrobicollis* adults were placed on each plant for approximately two weeks and were then removed. Plants were maintained in a growth chamber as described previously and number of adults emerging from each plant was recorded.

The addition of greenhouse soil mix to the top of pots resulted in an average of ten F1 adults per pot versus two adults per pot with the commercial rice hull mix. We now routinely add the 1:1:1:1 greenhouse soil mix to the top of the potting mixture. Adding a soil mix with a lower percentage of peat to the top of each pot, while sub-irrigating, could allow the larvae to pupate in drier, warmer soil. Larvae may also prefer the greenhouse soil mix for their pupal chambers rather than the peat-based potting mix.

Soil physical properties, temperature, and moisture level affect insect behavior in the soil (Villani and Wright 1990) as well as pupal survival (Lapointe and Shapiro 1999). Lapointe and Shapiro (1999) document an increase in pupal mortality in the citrus root weevil, *Diaprepes abbreviatus* L., at low or high soil moisture levels. Johnson et al. (2010) report that soil temperature as well as soil moisture affect clover root weevil, *Sitona lepidus* Gyllenhal, larval survival in soil. Our experiment was designed to develop a protocol to maximize the number of *C. scrobicollis* reared, not to address causes for differences in adult emergence between soil mixes. Future studies could examine the relationship among soil type, moisture level and temperature on adult emergence.

#### Experiment 2. Does transfer of *C. scrobicollis* from winter to spring conditions reduce development time?

In their native range, *C. scrobicollis* larvae exit from garlic mustard crowns in April and adults emerge from the soil from mid-May to late-June. In growth chambers, F1 adults emerged in simulated continuous winter conditions. To determine whether adults reared

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**Table 3. Duration of emergence of *Ceutorhynchus scrobicollis* F1 adults from potted garlic mustard plants when reared in growth chambers in a Biosafety Level 2 High Containment Facility. University of Minnesota, St. Paul. 2012.**

<table>
<thead>
<tr>
<th>Weeks after emergence of first F1 observed</th>
<th>Number of F1 adults (%)</th>
<th>Weekly F1 emergence (%)</th>
<th>Cumulative F1 emergence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>214</td>
<td>57.7</td>
<td>57.7</td>
</tr>
<tr>
<td>2</td>
<td>30</td>
<td>8.1</td>
<td>65.8</td>
</tr>
<tr>
<td>3</td>
<td>37</td>
<td>10.0</td>
<td>75.8</td>
</tr>
<tr>
<td>4</td>
<td>71</td>
<td>19.1</td>
<td>94.9</td>
</tr>
<tr>
<td>5</td>
<td>14</td>
<td>3.8</td>
<td>98.7</td>
</tr>
<tr>
<td>6</td>
<td>5</td>
<td>1.3</td>
<td>100.0</td>
</tr>
<tr>
<td>Total</td>
<td>371</td>
<td>100.0</td>
<td></td>
</tr>
</tbody>
</table>

*aWeekly emergence from 71 individual potted garlic mustard plants.*
in containment would emerge earlier when placed in winter/spring instead of continuous winter conditions, we conducted a study with two treatments. In the first treatment, caged plants with insects were placed into winter conditions in a growth chamber for 2 months followed by 2 months of spring conditions. For the second treatment, caged plants with insects were kept in continuous winter conditions for 4 months. Four to five pairs of weevils were added to plants and F-1 adults were reared as described previously. The experiment was replicated four times, with each replication consisting of one caged plant with weevils added.

When caged plants were transferred from winter to spring conditions, adults emerged approximately one week earlier than when kept in continuous winter conditions (data not shown). An average of seven adults per plant emerged when transferred to spring conditions compared to two adults when maintained in continuous winter conditions. Although not statistically significant (Number of F1 adults: df = 1, \(P = 0.28\); Days to F1 adult emergence: df = 1, \(P = 0.44\)), numerical trends indicate placement of caged plants into spring conditions, following two months of winter may increase the number of F1 adults and reduce the total F1 emergence time.

**Experiment 3. Effect of summer aestivation interval on fall oviposition.**

In Europe, *C. scrobicollis* adults emerge from early-May to mid-June, feed on garlic mustard plants for a short time, then aestivate during the summer. Adults become active in September and begin to lay eggs in mid-to-late-September (Gerber et al. 2009). We designed a study to determine the length of summer aestivation that induced females to deposit the greatest number of eggs in our simulated fall and winters when *C. scrobicollis* were reared in growth chambers.

Newly emerged F1 adults were placed onto garlic mustard plants, allowed to feed a minimum of two weeks in spring conditions (Table 1) and then placed into one of three summer aestivation treatments; three months (standard treatment), two months, or one month (Table 4). After completing the aestivation treatments, all caged plants were placed in fall conditions for one week, followed by winter conditions for three weeks. After the winter treatment, adults were removed from garlic mustard plants and placed into an oviposition test in winter conditions.

For the oviposition test, two females and one male (unless otherwise noted) were placed in a glass jar containing a garlic mustard leaf inserted into a piece of saturated florist foam and sealed with no-see-um cloth. After 2 to 3 days, leaves and petioles were dissected and checked for eggs. The number of eggs present per leaf was recorded. A minimum of four replications were completed, with each jar as a replication. Treatment means were separated with a Least Significant Difference test at the 0.05 level of significance.

After one month of summer aestivation, followed by 1 week of fall and three weeks of simulated winter conditions, all adults were feeding on plants, but only a total of three eggs were found out of five replications (Table 4). After two months of aestivation, adults were also actively feeding, but only two eggs were found out of five replications. Following the three-month aestivation period, a total of 69 eggs were found, an average of 13.8 eggs per leaf, a significantly higher number of eggs per leaf than the other aestivation periods (df = 3; \(P = 0.002\)). It should be noted that females laid a small number of eggs in an oviposition test after only two weeks spring treatment and without receiving an aestivation period.

**Table 4. Number of *Ceutorhynchus scrobicollis* eggs present in garlic mustard shoots after adults were placed in one, two or three month aestivation periods. Biosafety Level 2 Containment Facility. University of Minnesota, St. Paul, MN 2012.**

<table>
<thead>
<tr>
<th>Length of aestivation (months)</th>
<th>Length of fall/winter (weeks)</th>
<th>Total months</th>
<th>Number of eggs (total)</th>
<th>Number of eggs per shoot* (mean)</th>
<th>Feeding</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1/3</td>
<td>2</td>
<td>3</td>
<td>0.6</td>
<td>+</td>
</tr>
<tr>
<td>2</td>
<td>1/3</td>
<td>3</td>
<td>2</td>
<td>0.4</td>
<td>+</td>
</tr>
<tr>
<td>3</td>
<td>1/3</td>
<td>4</td>
<td>69</td>
<td>13.8</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>LDS (0.05)</td>
<td>2.1</td>
</tr>
</tbody>
</table>

*aWith no aestivation period, an average of 1.7 eggs per shoot were present, which did not differ from 1 or 2 months of aestivation.*
(1.7 eggs per leaf). From this we conclude that females are capable of depositing eggs after they emerge in the spring and prior to aestivation. In summary, the total number of eggs per leaf was highest with the standard three-month summer aestivation treatment, with a total length of time of four months before oviposition commenced (three months of aestivation followed by one week fall plus three weeks of winter).

**Experiment 4. Length of C. scrobicollis egg stage.** The possible presence of *P. consuetor* in field collected *C. scrobicollis* from Germany means that a minimum of one generation should be reared in a containment facility. This will ensure that any potential parasitoids can be eliminated prior to field release in North America. In our *C. scrobicollis* rearing protocol, we continually add mating pairs of adults to garlic mustard rosettes, then remove them after oviposition. *Perillus* endoparasitoids are known to attack adult or larval hosts but not eggs (Obrycki et al. 1985, Shaw 1988). Therefore, we wanted to isolate larvae from possibly parasitized adults to minimize the probability that *P. consuetor* could parasitize these larvae of the F1 generation. To accomplish this, it was critical to determine the length of the *C. scrobicollis* egg stage so that we could maximize the period of female oviposition while minimizing the risk of parasitoid attack to *C. scrobicollis* larvae or F1 adults.

To determine the length of time between *C. scrobicollis* oviposition and eclosion, *C. scrobicollis* adults were field collected near Berlin Germany in October 2018 and received into our containment facility. Plants and insects were maintained in a growth chamber simulating winter conditions of a 15/14 °C day/night temperature regime with a 9.5 h photoperiod (Table 1). This temperature regime was found to be optimum for *C. scrobicollis* oviposition (Gerber et al. 2002). After an acclimation period of approximately two weeks, four adult female and three-to-five male *C. scrobicollis* were placed in caged garlic mustard plants. Adults were added onto a total of eleven plants and the experiment was repeated in time.

After 48 hours, all adults were removed from plants. Starting at eight days after the adults were placed on garlic mustard plants, a single shoot was removed from five randomly selected plants, totalling 5 shoots per day. Shoots were dissected under a dissecting microscope and all eggs/larvae were counted, location on the petiole noted and egg/larvae development stage recorded. Shoots were sampled daily until 15 days after adults were placed onto plants. By this time, too few shoots remained on plants to continue the experiment and the majority of eggs had hatched.

There were no significant differences between the two trials of the experiment, so trials were combined (df = 1; *P* = 0.75). Initial eclosion was noted at 10 days after adults were placed on plants (Fig. 11). This means that eggs could have been 8 to 10 days old, since adults were removed from plants 48 hours after they were added. Since our goal is to minimize the probability that *P. consuetor* will parasitize larvae of the F1 generation, we will routinely remove adults from garlic mustard rosettes within 8 days after they are added, the point when eclosion was first observed. This will minimize the time when larvae are available for attack from *P. consuetor* if they happen to emerge from *C. scrobicollis* adults.

In containment, the majority, 56% of *C. scrobicollis* eggs were laid in the leaf petiole or at the base of the petiole while 44% of eggs were laid in the leaf blade. Similar results were found with larval location, 60% of larvae were located in the petiole or base of the petiole and 40% in the leaf blade, respectively. These results are similar to those reported by Gerber et al (2009) in field grown garlic mustard rosettes.

**Conclusions**

Ceutorhynchus scrobicollis can be successfully reared on caged garlic mustard plants in a growth chamber by alternating growth chamber temperatures and photoperiods to mimic natural conditions in its native range. In Germany, *C. scrobicollis* produces one generation per year and F1 adults emerge in late May. In containment, a new generation of adults emerged an average of 108 days after parent weevils were placed on plants. After emergence, F1 adults fed on garlic mustard rosettes for a minimum of two weeks before they were placed in a summer aestivation period. We found optimum oviposition after three-months of summer aestivation, followed with a week of fall, and three weeks of winter (four months total). In containment, we were able to produce a generation of *C. scrobicollis* every four months. However, the number of adults produced per plant was lower than that recorded for field reared plants at CABI in Delémont, Switzerland. Finally, we determined that the majority, 95% of potential F1 adults, were collected within the four week period after the first F1 adult appeared as described previously.

Lastly, the possible presence of the adult endoparasitoid, *P. consuetor*, in field collected adult *C. scrobicollis* from Europe will require that a minimum of one generation be reared in containment. This will
ensure the elimination of potential parasitoids prior to field release of *C. scrobicollis* in North America. Larvae first hatched 8 to 10 days after adults were placed on plants at 15/14°C day/night temperatures with a 9.5-hour photoperiod. In our rearing protocol, we will routinely remove *C. scrobicollis* adults from garlic mustard rosettes within 8 days after they are added. This will maximize the period of female oviposition while minimizing the time when larvae are available for attack from *P. consuetor*, if they happen to emerge from adults.

The rearing methods we have described provide baseline guidance for *C. scrobicollis* rearing in containment. We acknowledge that mass rearing procedures will need to be developed for conditions outside of containment, should *C. scrobicollis* be approved for field release in the U.S.

**Acknowledgments**

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Hidden Dangers to Researcher Safety While Sampling Freshwater Benthic Macroinvertebrates

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Abstract

This paper reviews hidden dangers that threaten the safety of freshwater (FW) researchers of benthic macroinvertebrates (BMIs). Six refereed journals containing 2,075 papers were reviewed for field research resulting in 505 FW BMI articles. However, danger was reported in only 18% of FW BMI papers. I discussed: 1) papers that did not warn of existing danger and consider researcher safety, 2) metric threshold values (e.g., chemical hazards), and non-metric dangers, (e.g., caves and aquatic habitats), 3), the frequency of danger occurrence, 4) baseline and extreme values. Examples of 28 danger factors that posed a threat to BMI researchers in water were compared by frequency per journal papers. FW dangers identified by metric thresholds present a safety limit not to be exceeded, whereas non-metric dangers do not have a threshold as further explained. Also, discussed was a recent thesis on civil engineering hydraulics that identified low-head dams as deceptive and an increasing source of drownings in 39 states. A safe shallow water maximum depth to wade and collect BMIs is proposed based on researcher height and gender, compared to human height means in a large database. Practical safety recommendations were presented to help protect the FW researcher avoid and survive hidden dangers.

Keywords: Researcher safety, danger factors, threshold values, accidents and drowning

The research of freshwater (FW) benthic macroinvertebrates (BMIs) can be rewarding and or dangerous. Aquatic field work is a required part of many BMI research projects and may include a wide variety and magnitude of risks (e.g., Dewailly et al. 1986, Howarth and Stone 1990, Courtenay et al. 2012, Orr 2017). In the above papers some researchers risked death, possibly due to inexperience, unknown equipment problems, scientific goals, etc. Some extreme examples of risk include deep dives (≥ 30 m) with scuba (Miyanishi et al. 2006), collecting BMIs in cave springs with high levels of toxic hydrogen sulfide (Tobler et al. 2006, 2013), accessing a deep cave with high CO2 levels, slippery vertical surfaces, rocky substrates, groundwater fed subterranean aquifers and springs (Howarth and Stone 1990), and crossing a swift, turbulent glacial stream for sediment samples (Orr 2017). Recently, the risk of drownings has been closely associated with large dams (Tschanz 2015), and especially at low-head dams in many streams and rivers (Hotchkiss et al. 2014, Kern 2014, Tschanz 2015). The sign in Fig. 1 advises everyone, including researchers, of danger if wading to collect BMIs near and below the large dam because of sudden fast discharge at various times to generate electric power. Fortunately, USA research scientists have a lower incidence rate (near 1%) for scuba dangers than other countries’ research scientists (Dardeau and McDonald 2007), who make more mistakes during deep dives at ≥ 30 m, e.g., Japanese divers (Miyanishi et al. 2006). According to Efrig (2017), a medical doctor who practices stress management prevention, most people hesitate to think about danger prevention until it is too late to make corrections. However, the discussion section explains what the FW BMI researcher should do to avoid unnecessary stress and dangers.

I evaluated papers in six peer-reviewed journals and other background literature on ecology of FW BMIs to determine if dangers were reported or a warning was included. I discovered that many papers sparsely informed the reader of potential dangers in FW BMI habitats. The danger features and safety concerns found became the objectives of this paper. The objectives were to: 1) find and discuss hidden-dangers in papers that did not warn of existing dangers in the field sampling process, 2) determine if those danger factors with metric threshold values (toxic and hazardous chemicals), non-metric...
dangers (e.g., caves with aquatic habitats), could be documented, 3) determine frequency of danger occurrence, 4) compare baseline values (safe) to above baseline and extreme (unsafe) values, 5) present practical and innovative safety recommendations to avoid dangers and fatalities prior to and during BMI surveys to enhance researcher safety. Examples of important government agencies that furnish water safety data and services to the public are presented here and in Table 1. The U.S. Environmental Protection Agency (USEPA) established the National Primary Drinking Water Regulations (also termed Standards) (USEPA 2017a). These Standards specify maximum contaminant levels (MCLs) with specific limits for drinking water contaminants to protect human health and are legally enforceable. Also, USEPA set Secondary Drinking Water regulations with secondary maximum contaminant levels (SMCLs) that are not federally enforceable or considered a threat to public health (USEPA 2017b). SMCLs are regulated mainly because of poor aesthetic water quality (e.g., metallic taste, odor, color, etc.). In addition to primary and secondary drinking water regulations, the Standards include states, U. S. Territories, and tribal lands (USEPA 2018). The USA drinking water and recreational water are also protected from pollution and degradation of discharges from municipal and industrial wastewater treatment plants into navigable water by the Clean Water Act (USEPA 1972). The U.S. Geological Survey (USGS 2017) researches water quality in states and U. S. Territories and reports stream flow and aquatic data in real-time from gaging stations by satellites. The Commission for Environmental Cooperation (CEC 2011) was a multi-year effort by Canada, Mexico, and the USA government agencies that developed and published North American Terrestrial Ecoregions—Level III. Each of the above countries described their ecoregions similarly by location, climate, vegetation, hydrology, terrain, wildlife, and land/human use. The regions relevant to this paper are listed in Table 1 concerning baselines and danger factors in USA Ecoregions (Wiken 2011a, b, c, d). Low precipitation included: a) Warm desert, b) Cold desert, c) Steepes, and d) as compared to High precipitation. The U.S. Centers for Disease Control and Prevention (USCDC 2015), sets pH, chlorine and other safety ranges. The U.S. Coast Guard (USCG 2017) presents annual statistics on water and boat related fatalities and accidents and issued the 2016 annual statistics report on inland waters (USCG 2017). This inland report compiles water statistics from data on rafts and boats and combines statistics with the states, U. S. Territories and media reports of recreational waters for related accidents, fatalities (that include drowning), and causes.

Fig. 1. Warning sign on approach to river below a spillway of large dam.
Table 1. Fourteen baseline metric values compared to exceeded metric values and extremes in freshwater ecosystems when collecting benthic macroinvertebrates as given in citations.

<table>
<thead>
<tr>
<th>Danger factors</th>
<th>Baseline metric values</th>
<th>References 1</th>
<th>Excessive metric values²</th>
<th>References¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Chemical hazards</td>
<td>Selenium (Se): 0.58 mg/l</td>
<td>USEPA 2017a</td>
<td>Se: 0.58 mg/l</td>
<td>Merriam et al. 2011</td>
</tr>
<tr>
<td></td>
<td>Iron (Fe): 0.3 mg/l</td>
<td>USEPA 2018</td>
<td>Fe: 3.0 mg/l</td>
<td>Lencioni et al. 2012</td>
</tr>
<tr>
<td></td>
<td>Nitrate NO₃: &lt; 10 mg/l</td>
<td>USEPA 2017a</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Sulfate SO₄: 1,725 mg/l</td>
<td>Davis 1980c</td>
<td></td>
<td>Davis 1980b</td>
</tr>
<tr>
<td></td>
<td>Dissolved oxygen:</td>
<td>USEPA 2018</td>
<td>O₂: 0.29 mg/l</td>
<td>Tobler et al. 2006</td>
</tr>
<tr>
<td></td>
<td>Class 1: &gt;9.5 mg/l</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Class 2: ≥ 8.0 mg/l</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2. Temperature (Hot)</td>
<td>25-28°C (77-82°F)</td>
<td>Olympic swimming pool 2019</td>
<td>54°C ~ Human scald point</td>
<td>PSEG 2017</td>
</tr>
<tr>
<td>(Cold)</td>
<td>In open water</td>
<td>Olympic open water 2019</td>
<td>0–14°C Extreme cold</td>
<td>NCCWS 2017</td>
</tr>
<tr>
<td></td>
<td>wetsuit required if &lt; 18°C</td>
<td></td>
<td>0–14°C Scuba dives</td>
<td>Renn and Evans 2012</td>
</tr>
<tr>
<td>3. Discharge</td>
<td>1.0 m³/sec</td>
<td>Gore 2006</td>
<td>≥ 937 m³/sec</td>
<td>Blinn et al. 1995</td>
</tr>
<tr>
<td>4. Maximum wade depth</td>
<td>0.4 m⁴</td>
<td>McDowell et al. 2008</td>
<td>~ 0.6 to 1.5 m</td>
<td>Barber and Minckley 1983</td>
</tr>
<tr>
<td>5. pH (Low)</td>
<td>6.5 neutral</td>
<td>USCDC 2015</td>
<td>pH: 3.2</td>
<td>Smucker and Morgan 2011</td>
</tr>
<tr>
<td>(High)</td>
<td>8.5 neutral</td>
<td>USCDC 2015</td>
<td>pH: 10.3</td>
<td>Davis 1980b</td>
</tr>
<tr>
<td>6. Low precipitation (Warm desert)</td>
<td>50-900 mm/yr</td>
<td>Wiken et al. 2011a</td>
<td>50 mm/yr Death Valley area, CA</td>
<td>Wiken et al. 2011a</td>
</tr>
<tr>
<td>(Cold desert)</td>
<td>800-2,000°C</td>
<td>Wiken et al. 2011b</td>
<td>100–150 mm/yr Tundra, n Northern Arctic</td>
<td>Wiken et al. 2011b</td>
</tr>
<tr>
<td>(Steepe)</td>
<td>518 mm/yr</td>
<td>Wiken et al. 2011c</td>
<td>300 mm/yr</td>
<td>Barber &amp; Minckley 1983</td>
</tr>
<tr>
<td>7. High precipitation</td>
<td>2,500 mm/y</td>
<td>Wiken et al. 2011d</td>
<td>3,397 mm/yr</td>
<td>Rosser &amp; Pearson 1995</td>
</tr>
<tr>
<td>8. Turbid</td>
<td>Maximum: 1 NTU/sample or any month 95% of samples &lt; 0.3 NTU</td>
<td>CCWP 2015</td>
<td>NTU: 450</td>
<td>O’Neil and Thorp 2011</td>
</tr>
<tr>
<td></td>
<td>NTU not to exceed:</td>
<td>USEPA 2017a</td>
<td>NTU: 450</td>
<td>Rosser and Pearson 1995</td>
</tr>
<tr>
<td></td>
<td>5.0 for non-conventional filtration and field</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>9. Gradient</td>
<td>≤ 4%</td>
<td>Charlebois and Lamberti 1996</td>
<td>33%</td>
<td>Wallace et al. 1995</td>
</tr>
</tbody>
</table>

(Continued on next page)
Many water accidents and drownings occurred in swift streams and rivers after spring rains and snow melt when discharge exceeded tolerance for aquatic researcher survival, especially when combined with low-head dams (Tschanz 2015). This seasonal trend occurred in states subject to intense precipitation in regional and mountain areas (Short and Ward 1980, Jacobi and Cary 1996), in fast tributaries of the Great Plains (Gray et al. 1983), and internationally such as the Shinano River in Japan (Kobayashi et al. 2013). Drownings also often occurred in streams and rivers and near low-head dams after spring melt through summer due to hidden hazards (Tschanz 2015). However, no accidents occurred at Alabama mill dams (low-head dams) in 20 streams with intact (unbroken), relic (broken and no spillway), and breached (some footings remained) low-head dams studied for fish assemblages in shallow water (mean depth ranged 0.16 – 0.50 m) (Helms et al. 2011). In a separate Alabama study of mollusks assemblages in 22 other small, low-head mill dams (height < 10 m), no accidents occurred by wading, snorkel, and scuba used in deeper sections (mean depth > 1.0 m) (Gangloff et al. 2011).

Materials and Methods

The selection of journals was partly from a list of 22 journals in FW aquatic ecology (Feminella and Hawkins 1995) and similar journals accessed mainly on-line, and the Colorado State University library.
The prioritized features were: 1) well-edited research papers, 2) papers recognized by indexing research engines (e.g., Web of Science and others), 3) papers on field ecology, FW BMIs including aquatic insect vectors, natural history, and conservation, and 4) papers from USA, regional, national, and international areas. The selected journals met each of the above priorities but varied by geographical emphasis of research in three areas: USA regionally, (e.g., Wiken et al. 2011a, b, c, d), nationally, and internationally). Journals selected that met the priorities were: Annual Review of Entomology (AREnt), Journal of Freshwater Ecology (JFWE), Journal of the North American Bentholological Society (DNABS) (renamed as Freshwater Science (FWS) in 2011), The Prairie Naturalist (PrNat), and The Southwestern Naturalist (SWAN).

Referenced journals, their citations, and prominent textbooks (e.g., Rosenberg and Resh 1993, Hauer and Lamberti 2006, Merritt et al. 2008, Thorp and Rogers 2015) were consulted.

I counted danger factors listed in Tables 2–4 by items per paper of journals, such as, lotic habitats (flowing waters) and lentic habitats (non-flowing waters), caves with aquatic habitats (Howarth 1983, Howarth and Stone 1990; James 2010; Tobler 2006, 2013), cold flowing waters (0–14°C) (Short and Ward 1980, Lencioni et al. 2012, National Center for Cold Water Safety (NCCWS 2017), and hot springs (few aquatic insects have adapted to exceed 39-42°C) (Bednarz 1979, Ward and Kondratieff 1992, Nolte et al. 1996, Alexander et al. 2011). Lotic water included discharge below dams and lentic water included marshes, lakes, and impoundments, e.g., reservoirs of dams. Large and small boats were counted with reference to size, currents, wave height, wind speed, and heavy and light water samplers. I counted tributaries that increased in discharge from precipitation and snow melt. Counts of dangerous large and low-head dams were accessed from an interactive database (Hotchkiss et al. 2014).

Shallow water samplers to study BMIs on substrate were mostly conducted with only a few of many types of available standard samplers e.g., Surber, to depths of 0.3 m, Hess to 0.5 m, D-frame to 0.3 m, kick net or Stanford-Hauer kick net from 0.2 to 1.0 m, (Surber 1937, Barbour et al. 1999, Hauer and Resh 2008, Merritt et al. 2008). No definitions were found in the literature on shallow water samplers related to human height or gender concerning wade depth limit for safety. However, a large statistical study in physical anthropology included age, race, ethnicity, gender, and height measurements (McDowell et al. 2008). A data summary of this work included: males 20 years and older: n = 4,482, \( \mu_{ht} = 176.3 \) cm, SE = 0.07; for females 20 years and older: n = 4, 857, \( \mu_{ht} = 162.2 \) cm, SE = 0.06. Here, I propose a method to apply the statistical data (McDowell et al. 2008) of height and means per gender to compare with researcher height and to obtain a safe shallow water wade depth. Also, I explain how to apply the means to prepare for safe wades. For example, the mean height for males was 1.76 m (or 176 cm in the above data summary) and if he works any of the standard samplers except the kick-net, due to his height he will be safe at 0.4 m. The same sampler depths safe for the male would be safe for the female at a mean ht of 1.62 m. However, for deeper kick-net wades up to 1.0 m, the male would be safe even if he stepped into a hole up to \( \sim 0.4 \) m (up to his hip in water). For the same female, her height would be unsafe (0.4 m for sampler + 0.4 m for a hole) and she would be in deeper water relative to the male at 0.8 m.

If river depth is unknown, the researcher should assess depth with numbered metric wading staff, or with a 1.0 m ruer, or consult USGS (2017) for \( \sim \) depth and velocity/river. The above samplers were designed for shallow streams and rivers with mostly cobble, gravel, and sand mixed substrate (e.g., Barbour et al. 1999, Hauer and Resh 2008, Merritt et al. 2008,) and were relatively uniform across channel width minus protruding objects (Gore 2006). Risk usually includes streams with combinations of dangers, e.g., collecting in a deep riffle (0.65 m) with swift currents (0.72–1.40 m/s) with substrate gravel and large cobble (Kobayashi
Table 2. Frequency of 28 freshwater danger factors, habitats, and event counts of safety concern to researchers in papers from reviewed journals with most total per journal given from top down.

<table>
<thead>
<tr>
<th>Danger factors with metric values and non-metrics</th>
<th>Number of Events in Journal Papers¹</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>PR</td>
</tr>
<tr>
<td>1. Lotic water habitats (e.g., flowing rivers,</td>
<td>38</td>
</tr>
<tr>
<td>streams, springs, and aquifers)</td>
<td></td>
</tr>
<tr>
<td>2. Unlisted dangers (e.g., shifting river</td>
<td>30</td>
</tr>
<tr>
<td>substrate sands)</td>
<td></td>
</tr>
<tr>
<td>3. Chemical water hazards (e.g., pesticides,</td>
<td>49</td>
</tr>
<tr>
<td>nutrients, gases)²</td>
<td></td>
</tr>
<tr>
<td>4. Impoundments</td>
<td>10</td>
</tr>
<tr>
<td>Dams</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>19</td>
</tr>
<tr>
<td>Reservoirs, lakes, pools</td>
<td>9</td>
</tr>
<tr>
<td>5. Temperatures of Cold</td>
<td>27</td>
</tr>
<tr>
<td>Cold ≤ 0–21°C &amp; Hot ≥ 40°C²b</td>
<td></td>
</tr>
<tr>
<td>Cold</td>
<td>27</td>
</tr>
<tr>
<td>Hot</td>
<td>0</td>
</tr>
<tr>
<td>6. Substrate hazards³</td>
<td>45</td>
</tr>
<tr>
<td>7. Discharge ~1 m³/s²</td>
<td>6</td>
</tr>
<tr>
<td>8. Dangerous animals and insect vectors</td>
<td>2</td>
</tr>
<tr>
<td>9. Exceeded maximum depth of 0.4 m²²</td>
<td>17</td>
</tr>
<tr>
<td>10. Boats, cables, grabs, nets, seines³</td>
<td>45</td>
</tr>
<tr>
<td>11. Floods &amp; Flash floods per study</td>
<td>18</td>
</tr>
<tr>
<td>12. Total pH ≤ 6.5 – ≥ 8.5²</td>
<td>8</td>
</tr>
<tr>
<td>High</td>
<td>8</td>
</tr>
<tr>
<td>Low</td>
<td>0</td>
</tr>
<tr>
<td>13. Low precipitation ≤ 500 mm/y</td>
<td>3</td>
</tr>
<tr>
<td>(e.g., deserts, Steppes, Tundra)²</td>
<td></td>
</tr>
<tr>
<td>14. High precipitation &gt; 2500 mm/yr²</td>
<td>3</td>
</tr>
<tr>
<td>15. Marsh, swamp, bog</td>
<td>6</td>
</tr>
<tr>
<td>16. Night &amp; twilight in aquatic habitats</td>
<td>17</td>
</tr>
<tr>
<td>17. Turbid water ≥ 5 NTUₘ²</td>
<td>12</td>
</tr>
<tr>
<td>18. SCUBA/Snorkel swims, dives with</td>
<td></td>
</tr>
<tr>
<td>wide range of depths, time underwater,</td>
<td></td>
</tr>
<tr>
<td>discharge, distance, and temperature</td>
<td>0</td>
</tr>
<tr>
<td>Gradient ≥ 4 % m²/km²</td>
<td>0</td>
</tr>
<tr>
<td>20. Sewage ≥ USEPA test limit of total/fecal</td>
<td>0</td>
</tr>
<tr>
<td>coliform/MPN, BOD²</td>
<td></td>
</tr>
<tr>
<td>21. Conductivity 400 µS/cm²</td>
<td>9</td>
</tr>
<tr>
<td>22. Caves with aquatic habitats (e.g.,</td>
<td>0</td>
</tr>
<tr>
<td>streams, waterfalls, &amp; seeps)</td>
<td></td>
</tr>
<tr>
<td>23. High physical effort (e.g., collect near</td>
<td>7</td>
</tr>
<tr>
<td>hazardous dams/swift tailwaters, in deep</td>
<td></td>
</tr>
<tr>
<td>sinkholes)</td>
<td></td>
</tr>
<tr>
<td>24. High winds &gt; 3-5 m/s (8-12 mph)²</td>
<td>16</td>
</tr>
<tr>
<td>25. High altitude ≥ 2,425 m (8000 ft)²</td>
<td>0</td>
</tr>
<tr>
<td>26. Many sampling sites ≥ 40</td>
<td>1</td>
</tr>
<tr>
<td>27. TDS ≥ 500 mg/²</td>
<td>0</td>
</tr>
<tr>
<td>28. Waterfalls</td>
<td>0</td>
</tr>
</tbody>
</table>

¹ Abbreviations from complete journal titles given in Table 4.
² Exceeded USEPA primary contaminant level or metric baseline safety limit from authorities in citations.
³ Substrate physical hazards: Sharp metal, glass, ice, rocks, holes, roots, snags, algae with mucus, stumps, vascular plants, etc.
⁴ Boats: small, e.g., canoe in swift currents, waves to 0.6 m; large boats with large net, grabs to trawl deep water, e.g., ≥ 21 m, waves to 4 m.
et al. 2013), and increases in cold water with a slippery substrate (Wellnitz et al. 1996).

Reviewed papers were excluded if: 1) they were ambiguous, 2) BMIs were not resolved at least to family to be of value as a bioindicator and for BMI vector surveys (Rosenberg and Resh 1993), 3) BMIs were not identified from a natural, FW habitat by location and date, 4) the author or co-author never entered the water in a survey of a FW habitat, 5) BMIs were < 1.0 mm, 6) study was speculative, 7) specimens were petrified fossils, 8) data were not original. Two papers (Hotchkiss et al. 2014, Kern 2014) were exceptions to above exclusions in this review because of a timely civil engineering thesis on low-head dam dangers, an interactive database and practical solutions to prevent accidents in drownings.

If authors indicated moderate or high turbidity, swift discharge, or slippery waterfalls, their professional judgement was accepted. Aircraft were considered equivalent to boats in deep, swift water Paragamian (2010) as potentially dangerous. Helicopters and a few fixed winged aircraft that were deployed in early eradication projects over African fast streams and rivers (Davies et al. 1962), with pesticides applied at spillways of large dams and in rapids (Davies 1994) were included. Helicopters utilized in difficult to access locations such as the Mackenzie River, its tributaries, and Canadian wetlands (Scott et al. 2011) were also included.

The term, unlisted dangers, is defined here as an infrequently occurring and non-metric danger (e.g., sinkhole at cave entrance, Howarth 1973), collections below high hazard dam (Davis 1980b), runaway barges on large rivers and shifting bottom sands (Way et al. 1995).

In Table 3, non-metric danger factors were not ranked because they vary with conditions, including slippery substrate, thin ice over streams and ponds, obscured deep holes in rivers, underwater snags, stumps, and unique events (e.g., a cave passageway with a surprise flood or rocks/ide). Other dangers possibly overlooked by researchers: barbed wire on substrate of shallow, turbid streams, sloughs, and lakes (RDS unpublished).

Table 3 presents examples of a safe stream with few dangers in the Devil's River, Texas --- a clear, riffle bearing stream with springs, intermittent pools, low waterfalls, limestone substrate, excellent water quality, and high BMI diversity (Davis 1980c). In Table 1, baseline studies were presented first with safe low metric values and compared to metric highs and extremes as examples of contrast in collection safety.

Open the following web address (at the BYU website) to access the USA low-head dam interactive database (Hotchkiss et al. 2014) with a color map of the states showing one or more fatalities at these structures http://krcproject.groups.et.byu.net/browse.php The reader can see the total fatalities recorded was 555 from 276 sites. This information was found by accessing the above website on 19 December 2018, as shown on the colored map of 39 state locations with submerged fatality points at intact dams. New incidents may be reported by clicking the tab on top of the color map page (Hotchkiss et al. 2014).

Results

Six refereed journals in Table 4 containing 2,075 papers were reviewed for field research resulting in 505 FW BMI papers and of these, 265 (52%) contained danger. However, the above 505 journal papers reported only 90 (18%) with danger.

Extreme high and low frequency totals per journal were reported in Table 2. The most reported frequency total for combined journals with danger that exceeded other metric threshold values was lotic waters with a total of 1,243. The most common non-metric danger near the top of Table 2 with most reported dangers was unlisted dangers with a total of 987. Of the lows with three reported zeroes per journal, TDS (metric) included a total of 40. The least reported low per journal was waterfalls (non-metric) with a total of only 29. Each of these extreme highs and lows were explained in the discussion.

Of the 14 metric dangers listed in Table 2, only 12 represent one metric danger value. However, when the two other danger factors, were included (e.g., chemical water hazards and sewage effluents), they account for the total 14 metric values. Sewage involved several required microbiological and chemical tests to express their values (e.g., total and fecal coliform counts per sample and biological oxygen demand (BOD₅) in mg/l). For a complete list of chemicals with safety thresholds for drinking water quality, the Standards should be consulted in addition to examples in Table 1, for any of the above test groups.

The 14 non-metric danger factors may be considered on a large scale with examples reported by specific location and other features. Some examples were: low-head dams with a history of multiple fatalities, unmanaged old dams, remote caves that flood, deep caves with difficult access and exit, and remote waterfalls especially slippery during high seasonal flow (Holzenthal 1995).
In 2016 the US Coast Guard reported 437 drownings for recreational boats and 10 drownings at dams and locks in states and U.S. Territories, but did not mention low-head dams. However, low-head dams have recently been recognized as a public danger, mainly due to the Kern thesis and were included in this paper because of the high drowning rate and increase in fatality postings on the interactive database. Recent statistics from the interactive database with 89 photos of dams (Hotchkiss et al. 2014) were accessed on 19 December 2018 by RDS. This database indicated that low-head dams in 39 states accounted for 555 drownings fatalities, the number of fatality sites was 276, and had a maximum of 12 fatalities at a single site. Photos of 84 intact low-head dams were on streams and rivers, and photos of 15 large dams with locks were on rivers in the above database where drownings occurred.

Discussion

To my knowledge this may be the first paper to find, document, and discuss danger factors were mostly unreported in the FW ecological research and imperiled researcher safety. This paper originated after an inquiry to determine if six peer reviewed journals reported the occurrence of danger together with their subject matter in FW ecological field research of BMIs. This inquiry led to the objectives of this paper with some new and surprising results and recommendations to enhance FW researcher safety. Beginning here is an overview of five objectives that confirm 28 dangers were found.

In objective one, hidden dangers occurred, but few (18%) were reported or presented a warning to the reader. However, 52% of the journal papers contained dangers (Table 4). The sparse journal information on unreported danger implies that FW researchers may have depended on swimming ability, experience, and other information to avoid danger. However, some researchers are: unable to swim, inexperienced, or had minimal safety training, and some dangers are hidden and difficult to anticipate or have a deceptive calm appearance (e.g., low-head dams). Examples of the danger factors involved were from a wide range of sources in addition to the journal papers such as prominent textbooks (Hauer and Lamberti 2006, Merritt et al. 2008, Thorp and Rogers 2015), and research scientists in the Literature Cited section (Howarth 1983, Lencioni et al. 2012, Mebane et al. 2012, Kern 2014, and many others).

The image of a warning sign in the Introduction illustrated how some states warn public waders of swift currents and rising water level danger below hydroelectric dams. Some states advise of danger on-line and post warning signs to boaters upstream of low-head dam hazards at portals to prevent fatalities.

In objective two, hidden dangers caused accidents and especially drownings fatalities that were traced to metric threshold values (e.g., chemical hazards) and non-metric dangers (e.g. caves with FW habitats and or night collections in water) from field results and cited papers. However, only one mention of a scientific researcher was found as an example of extreme risk in deep (dives > 30 m) (Miyanishi et al. 2006). Some other examples of taking high risks include investigations in remote, deep caves with water hazards (Howarth 1973) and toxic gases (Howarth and Stone 1990, James 2010), deep arctic lakes (Luoto et al. 2013), desert sinkholes (Bednarz 1979, Macanowicz et al. 2013). Hidden dangers that involved fatalities were reported mainly at intake points upstream near large dams and below the dam release points for high discharge near and downstream of spillways, but most drownings occurred at low-head dams (Tschanz 2015). Videos and many photos of low-head dams and larger dams identified danger and drowning points seen on an interactive database and map of 39 states was revealed by the thesis in civil engineer hydraulic (Kern 2014). One of the most inconspicuous and deadly hidden dangers is low-head dams. Waders that venture on the dam and swimmers and boaters, canoeists, and kayakers that flow over the dam were found trapped in a strong reverse backflow currents and in turbulence near and downstream of the dam according to Kern (2014) and Tschanz (2015).

Several danger factors such as scuba and caves with FW BMI habitats may be metric or non-metric dangers depending on danger intensity, e.g., zero oxygen level in a scuba air tank if in deep water or in a deep cave. However, scuba dives in deep water were few and the exception. Also, the large caves of Mulu (Indonesia) and Undara (Australia) had challenges minimized because of carefully planned expeditions with experienced cave scientists, new and more precise technological equipment to map and report data, and required permits that specify conditions to explore, collect specimens, and include regulated time limits by country officials.

Data that supports important conclusions on hidden dangers originated in Table 4. Three examples show that Table 4 presents a wide range of data from the six reviewed journals but, requires some interpretation. For example, on inspection of row 3, column 3, it shows the journal with the
Table 3. A list of the 14 non-metric danger factors identified with cited references, examples, and locations.

<table>
<thead>
<tr>
<th>Non-metric danger factors</th>
<th>References</th>
<th>Danger factor</th>
<th>Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Lotic water habitats, e.g., river, streams, and springs (Partly metric)</td>
<td>Blinn et al. 1995</td>
<td>Collected benthos in substrate below large dam in fast current and 25 km downstream at 6.8 m depth by snorkel and scuba</td>
<td>Colorado River at Glen Canyon Dam and Lees Ferry, AZ</td>
</tr>
<tr>
<td>Naiman 1979</td>
<td></td>
<td>Death Valley drying spring with slow flow</td>
<td>Near Tecopa, CA</td>
</tr>
<tr>
<td>2. Unlisted dangers</td>
<td>Way et al. 1995</td>
<td>Collecting in river, with heavy barges, and in shifting river sands to set and retrieve BMI underwater traps by scuba</td>
<td>Marshalls Point Mississippi River, MS</td>
</tr>
<tr>
<td>Macanowicz et al. 2013</td>
<td></td>
<td>Accessed sinkhole with groundwater and steep sides (90° angle)</td>
<td>Bitter lake, NWLR, NM</td>
</tr>
<tr>
<td>3. Impoundments e.g., Dams and lentic waters e.g., reservoirs, lakes and pools</td>
<td>Adams 2011</td>
<td>Collect crayfish down-stream and near old, remote, unstaffed dams and impoundments</td>
<td>Upper Little Tallahatchie River, sub-basin, northern MS</td>
</tr>
<tr>
<td>Davis 1980b</td>
<td></td>
<td>High hazard dam seeping saline water</td>
<td>The upper Pecos River, TX</td>
</tr>
<tr>
<td>Kern 2014</td>
<td></td>
<td>Author developed interactive database of fatalities at low-head dams in USA states with details and many photos</td>
<td>See authors database under Hotchkiss et al., 2014 in literature cited and click on hypertext</td>
</tr>
<tr>
<td>4. Substrate/physical water hazards(^1)</td>
<td>Helms et al. 2011</td>
<td>Relic low-head mill dams with scattered concrete pieces mixed in substrate</td>
<td>20 old mill river sites in AL</td>
</tr>
<tr>
<td>Wellnitz et al. 1996</td>
<td></td>
<td>Slippery rocks on biofilm</td>
<td>High altitude stream, St. Louis Creek, Rocky Mt. National Park, CO</td>
</tr>
<tr>
<td>5. Dangerous animals and insect vectors</td>
<td>Davies et al. 1992, Davies 1994</td>
<td>Vector control of <em>Simulium damnosum</em> complex an important vector of onchocerciasis or river blindness disease</td>
<td>Rivers, falls, rapids of East and central Africa</td>
</tr>
<tr>
<td>Takken and Knoles 1999</td>
<td></td>
<td>Surveys of <em>Anopheles gambiae</em> complex and important malaria vector</td>
<td>Sub-Sahara, Africa and Arabian Peninsula</td>
</tr>
<tr>
<td>Townsend et al. 2012</td>
<td></td>
<td>Crocodile slide at field site</td>
<td>Darby River, tropical Australia Hannacroix Creek, a tributary to Hudson River, NY</td>
</tr>
<tr>
<td>Waidt et al. 2013</td>
<td></td>
<td>Electric eels collected in mud flats and marsh with electro-shocker, and in float traps at night to 2 m depth</td>
<td></td>
</tr>
<tr>
<td>6. Boat size, wave heights, nets, sampling, grabs, etc.(^2)</td>
<td>Macanowicz et al. 2013</td>
<td>Enter desert sinkholes, walls top to bottom at a steep angle</td>
<td>Bitter Lake NWLR, NM</td>
</tr>
<tr>
<td>Schoenebeck and Brown 2010</td>
<td></td>
<td>Night collection by boat with heavy sampling equipment, e.g., grabs, cables, and long nets</td>
<td>Lake Cochran and Lake Madison, SD</td>
</tr>
<tr>
<td>Way et al. 1995</td>
<td></td>
<td>Boat, cables, and winch to guide concrete blocks by scuba, on river substrate</td>
<td>Marshall’s Point, Mississippi River, MS</td>
</tr>
</tbody>
</table>

(Continued on next page)
Table 3. Continued.

<table>
<thead>
<tr>
<th>Non-metric danger factors</th>
<th>References</th>
<th>Danger factor</th>
<th>Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>7. Floods and Flash floods</td>
<td>Barber and Minckley 1966, 1983</td>
<td>Sonoran Desert flash floods at macrobenthic stream sites</td>
<td>Araviaca Creek, AZ</td>
</tr>
<tr>
<td></td>
<td>Fisher 2011</td>
<td>Missouri and Yellowstone rivers flood rate studies</td>
<td>McKenzie Co., ND</td>
</tr>
<tr>
<td></td>
<td>Gray 1983</td>
<td>Spring flash floods from snow melt in headwater streams of collection sites</td>
<td>Piceance Basin, CO</td>
</tr>
<tr>
<td></td>
<td>Howarth 1973</td>
<td>Danger occurs when lava cracks release floods from surface rain and irrigation water into passageway</td>
<td>Lava tube on Island of Kauai, HI</td>
</tr>
<tr>
<td>8. Marsh, swamp, bog</td>
<td>Batzer and Wissinger 1996</td>
<td>Study of population and community ecology in large wetlands including insect vectors</td>
<td>From Newfoundland bogs to Everglades, FL</td>
</tr>
<tr>
<td>9. Night and twilight in aquatic habitats</td>
<td>Fisher 2011</td>
<td>Night micro-crustacean surveys at seasonal river pulse</td>
<td>Missouri and Yellowstone rivers in northwest ND</td>
</tr>
<tr>
<td></td>
<td>Kershner and Lodge 1995</td>
<td>Night crayfish census</td>
<td>Northern Lakes, WI</td>
</tr>
<tr>
<td>10. Scuba/snorkel dives (^3)</td>
<td>Hovarth et al. 1996</td>
<td>Snorkel-scuba dives: 6 lakes and 38 streams to find zebra mussels dispersal and populations</td>
<td>St Joseph R., IN and MI</td>
</tr>
<tr>
<td></td>
<td>Vaughn and Taylor 1999</td>
<td>Snorkel and scuba dives in reservoirs and river census of clams for ecological status</td>
<td>Little River (tributary), dam areas, and main stem of Red River, OK</td>
</tr>
<tr>
<td></td>
<td>Wisniewski et al. 2013</td>
<td>Strenuous snorkel dives against current to search for glochidia on rare fish in sharp rock crevices</td>
<td>Flint River, GA</td>
</tr>
<tr>
<td>11. Caves and their aquatic habitats (Partly metric event)</td>
<td>Howarth 1983 (cite #64)</td>
<td>Pioneering study of troglobites in 50 lava tubes</td>
<td>Kauai, Hawaii, Maui, Oahu Islands, HI</td>
</tr>
<tr>
<td></td>
<td>Howarth and Stone 1990</td>
<td>First troglobite arthropod community study in deep cave with high humidity, high CO2, level 200% more than ambient air outside entrance and zero level O2 at 830 m inside.</td>
<td>Bayliss is largest cave of Undara volcano lava tube complex, Queensland, Australia</td>
</tr>
<tr>
<td>12. High physical effort</td>
<td>Charlebois and Lamberti 1996</td>
<td>Extensive snorkeling and swims to monitor effect of invasive crayfish consumers of BLMs and periphyton</td>
<td>Middle Branch, Ontonagon River, MI</td>
</tr>
<tr>
<td></td>
<td>Fisher 2011</td>
<td>Missouri and Yellowstone rivers flood rate studies</td>
<td>McKenzie Co., ND</td>
</tr>
<tr>
<td></td>
<td>Ozersky et al. 2011</td>
<td>16-year Scuba monitoring at bottom of Canadian cold lake for zebra mussel ecology</td>
<td>Lake Simcoe, Canada</td>
</tr>
<tr>
<td>13. Many sampling sites ≥ 40</td>
<td>Larsen and Olden 2013</td>
<td>Conducted crayfish census at 100 lake sites.</td>
<td>Puget Sound lowlands, WA</td>
</tr>
<tr>
<td>14. Waterfalls</td>
<td>Charlebois and Lamberti 1996</td>
<td>Bond Falls</td>
<td>Ontonagon River, MI</td>
</tr>
</tbody>
</table>

\(^1\) See list of substrate physical hazards at bottom of Table 2
\(^2\) See list of boat size, wave heights, nets, sampling grabs, etc. at bottom of Table 2 and effects of wind velocity on boats in Table 1.
\(^3\) See Discussion for explanation.
most papers was the second lowest in aquatic dangers (column 7). However, row 2, column 3 also shows that the journal with the lowest total papers surprisingly correlates with row 2, column 8 at 83% for the most danger papers per journal. This surprise result was because almost every paper submitted to the PrNat in the second case contained at least three dangers. However, the total number of dangers was gleaned from journal papers that required time consuming reads of text, tables, figures, and citations.

This paper included two innovative contributions from researchers. First the M.S. thesis below that identified dangers associated with low-head dams and presented solutions to correct this problem (Kern 2014). Second, an innovation (proposed by RDS) herein that FW researchers may use to evaluate their height by gender to means from a large database (McDowell et al. 2008) to prepare for safe wading when considering substrate holes and stream conditions. Researchers should know bottom depth and the conditions (e.g., substrate composition, water clarity, and current velocity). It is important to find wade depths up to 1.0 m so researchers know their height is ample for water depth and holes to 0.4 m depth or deeper before sampling substrate for FW MBIs.

In objective three, dangers were considered by frequency of occurrence. The frequency of occurrence for reviewed journals (Table 2) with dangers is explained here. Lotic waters were the most frequently reported metric partly because of many tributaries to streams and rivers and had the most citations of reported papers with danger (however, many were short term or less than a one-year study). Unlisted danger was the most reported non-metric with high frequency. Infrequent kinds of danger were experienced, sometimes resulting in a variety of incidents at one location (e.g., shifting bottom sands, and variable currents when scuba diving to place artificial substrates

### Table 4. Six journals reviewed for danger during collection of freshwater benthic macroinvertebrates.

<table>
<thead>
<tr>
<th>Journal</th>
<th>Years Reviewed</th>
<th>Total Papers Reviewed</th>
<th>Total Freshwater BMI Papers</th>
<th>BMI Papers with Implied Danger</th>
<th>Percent with Implied Danger</th>
<th>BMI Papers with Reviewed Danger</th>
<th>Percent with Reviewed Danger</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>2011–2013</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>The Prairie Naturalist</td>
<td>2005–2013</td>
<td>175</td>
<td>18</td>
<td>6</td>
<td>33</td>
<td>15</td>
<td>83</td>
</tr>
<tr>
<td>PrNat</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>The Southwestern Naturalist</td>
<td>1979–1984</td>
<td>773</td>
<td>71</td>
<td>14</td>
<td>20</td>
<td>37</td>
<td>52</td>
</tr>
<tr>
<td>SWAN</td>
<td>2012–2013</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>JFWEcol</td>
<td>2, 3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>JNABS</td>
<td>2011</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Freshwater Science</td>
<td>2012–2013</td>
<td>204</td>
<td>111</td>
<td>13</td>
<td>12</td>
<td>53</td>
<td>48</td>
</tr>
<tr>
<td>FWS</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Totals</td>
<td></td>
<td>46</td>
<td>2,075</td>
<td>505</td>
<td>90</td>
<td>18</td>
<td>265</td>
</tr>
</tbody>
</table>

1 Abbreviation of BMI = benthic macroinvertebrate for this table.

2 Journal abbreviations apply to Tables 2 and 4.

3 In 2011 the Journal of the North American Benthological Society (JNABS) changed its name to Freshwater Science (FWS).
on river bottom during heavy barge traffic, deep scuba dives (≥ 30 m) (Miyanishi et al. 2006), wading across swift glacial streams (Orr 2017), collecting FW BMIs at or below hazardous, old, unattended, and abandoned dams (Davis 1980b, Adams 2013, Tschanz 2015). TDS reported the most zero values per journal and second lowest frequency. Many papers were on specifics unrelated to TDS (e.g., surveys for endangered species, bioindicators of water quality, insect vectors, and biocontrol releases). However, TDS may have many origins in springs, streams, and rivers (e.g., groundwater with nitrates, and limestone aquifers, excessive turbidity, and sewage) (Davis 1980a, b, c). Hot springs (metric) and caves with aquatic habitats (non-metric) were reported as tied because they had two zero values per journal. Few papers reported BMIs (especially insects) in hot springs if water exceeded 39-42°C because of the lethal effect to most FW BMIs (Table 1, item 2). AREnt and SWAN exceeded four other journals in reporting a combined total of 81 dangers in aquatic habitats of caves as a result of international surveys, including some Pacific island caves. Last reported were waterfalls because they had the fewest reported total frequency (non-metric) of any danger factor per journal. However, high waterfalls have common dangers that may lead to fatalities mainly because of slippery rocks in wet season (Holzenthal 1995) and may have treacherous, sharp rocks in dry or cold season, and require high elevation climbs. (Finn and Poff 2011).

In objective four, two danger groups considered equally dangerous for field safety, are explained here. The 14 non-metric dangers were identified as not having specific measurable values and may not be normalized because they are unique and have separate natural habitats such as caves, bogs, marshes, swamps, etc., (Table 3). Also, the non-metric dangers are characterized by their collective features (e.g., floods) and events (e.g., twilight). The 14 metrics included measurable baseline threshold values having maximum safety limits (or minimum for low temperature, precipitation, and pH) or if extended beyond baseline values they are unsafe as compared in Table 1 (e.g., MCL or if extended beyond baseline values they are unsafe as compared in Table 1 (e.g., MCL)

Acknowledgments
I thank Dr. Boris C. Kondratieff, Colorado State University for early manuscript advice and for sharing collecting experiences on water safety during his many stream surveys in the USA and worldwide. I thank Pat Fritts, Director, Arkansas Game and Fish Commission for approval of copyright license.
number 8052-1177 to display the warning sign image of danger to waders that swift waters may suddenly rise below a large dam spillway. I thank Randal Owens, US Fish and Wildlife Service biologist (retired) for sharing experiences during his many years of ecological research on the Great Lakes in deep water, storms, snow, ice, and night trawls. Helpful insights from anonymous reviewers that improved the paper were essential and appreciated. I remain thankful to an unknown alert fisherman that threw me a rope and ring in the cold, swift current of the Trinity River north of Dallas-Ft. Worth in my early collecting days.

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Davis, J. R. 1980c. Species composition and diversity of benthic macroinvertebrates of...


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Effects of Laboratory Heat Stress on Mortality and Web Mass of the Common House Spider, Parasteatoda tepidariorum (Koch 1841) (Araneae: Theridiidae)

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Abstract

We determined the effects of laboratory heat stress on web construction of Parasteatoda tepidariorum (Koch) (Araneae: Theridiidae) by measuring the survival and web mass of specimens after a 48-h period within a temperature chamber at 21, 30, 35, 40, or 50°C. The 21, 30, and 35°C treatments had the highest mean survival rate (100%), the 50°C treatment had the lowest (0%), and the 40°C treatment was intermediate (58%). The 21, 30, and 35°C treatments had the highest mean web mass, and the 40 and 50°C treatments had the lowest. Web mass did not correlate with spider mass for specimens across all temperature treatments. While acclimation temperature and humidity fluctuated throughout the 3 weeks of the study, neither variable affected web mass. This study demonstrates the sublethal effect of temperature on web construction, an effect that may ultimately be lethal in nature if a spider was unable to construct its web.

Keywords: Spider, Parasteatoda tepidariorum, heat, stress, sublethal, web

Ambient temperature is widely known to have many important effects on spiders and other organisms. Because spiders are ectothermic, their metabolic rate and subsequent activity level is directly tied to temperature. Moreover, because many spiders must engage in a specific physiological behavior—web construction—in order to feed and mate, any temperature that affects this behavior will be ultimately be lethal, even if that temperature does not directly cause death.

Despite the importance of spiders in controlling pest insects in both natural and agricultural ecosystems (Nyffeler and Benz 1987, Uetz et al. 1999), the study of spider thermal ecology has historically gotten little attention. As of the last comprehensive review, thermal tolerance values were known for <0.1% of species (Humphreys 1987). Several studies, however, have shown that spiders actively seek out habitats with preferred temperatures, and may position their web to optimize sun exposure and temperature within a habitat (Biere and Uetz 1981, Lubin et al. 1991, Lubin et al. 1993). Spider species found in relatively cool natural environments tend to mature at a slower rate when exposed to high temperatures in the laboratory. The reverse is true for spiders that inhabit warm environments when exposed to colder temperatures (Li and Jackson 1996). Due to concerns about anthropogenic temperature increase, and the importance of spiders in both natural and agricultural systems, it is necessary to study the effects of high temperatures on spiders, especially subtle effects on web construction.

The common house spider, Parasteatoda tepidariorum (Koch 1841) (Araneae: Theridiidae) is thought to be native to South America and is currently established throughout much of the world, where it is associated with both natural environments and human dwellings (Edwards 2001). The species builds a tangle-style web that houses both males and females, making it crucial for feeding and reproduction. House spiders are increasingly being used as model organisms for genetic, embryological development, and evolutionary studies (McGregor et al. 2008, Mittmann and Wolff 2012, Posenien et al. 2014, Schomburg et al. 2015). Tanaka (1989, 1991, 1992) determined its life cycle in Japan, as well as the effects of temperature and photoperiod on its maturation and reproduction.

Barghusen et al. (1997) studied the effects of laboratory temperatures between 5–30°C on P. tepidariorum web construction over 6-d periods and found the highest web mass was produced by specimens housed at 20°C, although there was statistical overlap between groups housed between 15–30°C. That study did not test spider mortality and, thus, utilized fairly low temperatures.

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Despite the importance of spiders in controlling pest insects in both natural and agricultural ecosystems (Nyffeler and Benz 1987, Uetz et al. 1999), the study of spider thermal ecology has historically gotten little attention. As of the last comprehensive review, thermal tolerance values were known for <0.1% of species (Humphreys 1987). Several studies, however, have shown that spiders actively seek out habitats with preferred temperatures, and may position their web to optimize sun exposure and temperature within a habitat (Biere and Uetz 1981, Lubin et al. 1991, Lubin et al. 1993). Spider species found in relatively cool natural environments tend to mature at a slower rate when exposed to high temperatures in the laboratory. The reverse is true for spiders that inhabit warm environments when exposed to colder temperatures (Li and Jackson 1996). Due to concerns about anthropogenic temperature increase, and the importance of spiders in both natural and agricultural systems, it is necessary to study the effects of high temperatures on spiders, especially subtle effects on web construction.

The common house spider, Parasteatoda tepidariorum (Koch 1841) (Araneae: Theridiidae) is thought to be native to South America and is currently established throughout much of the world, where it is associated with both natural environments and human dwellings (Edwards 2001). The species builds a tangle-style web that houses both males and females, making it crucial for feeding and reproduction. House spiders are increasingly being used as model organisms for genetic, embryological development, and evolutionary studies (McGregor et al. 2008, Mittmann and Wolff 2012, Posenien et al. 2014, Schomburg et al. 2015). Tanaka (1989, 1991, 1992) determined its life cycle in Japan, as well as the effects of temperature and photoperiod on its maturation and reproduction.

Barghusen et al. (1997) studied the effects of laboratory temperatures between 5–30°C on P. tepidariorum web construction over 6-d periods and found the highest web mass was produced by specimens housed at 20°C, although there was statistical overlap between groups housed between 15–30°C. That study did not test spider mortality and, thus, utilized fairly low temperatures.
The objective of the current study was to compare both mortality and web mass of *P. tepidariorum* based on temperatures warmer than those used by Barghusen et al. (1997).

### Material and Methods

Adult female specimens of *P. tepidariorum* were obtained from Spider Pharm, Yarnell, AZ (https://spiderpharm.com/) in two batches; the first batch encompassed the first two rounds of trials and the second batch was the third round. Prior to trials, each specimen was maintained in the lab in separate 8 cm H × 7 cm D round clear plastic jars and exposed to ambient photoperiod and indoor temperature. Room temperature (°C) and relative humidity (%) were measured at 9:00 am and 6:00 pm each day and the mean between them recorded. Each specimen jar included 3 round wooden dowels (~5.0 cm L × 2.0 cm W × 0.3 cm D), around which the spiders could spin webs. Specimens were not fed during the holding period except for a single muscid fly adult—either included with the Spider Pharm order or wild-caught—which was offered 24 hours before that specimen’s trial. All spiders caught and wrapped their prey, although we could not determine how much of that prey was consumed.

Three rounds of temperature trials on *P. tepidariorum* were run during the period of mid-May to mid-June 2018 (Table 1). Each round consisted of 5 different temperature treatments: 21, 30, 35, 40 and 50°C. We assumed that 50°C would be lethal to the spiders, whereas the 21°C treatment approximated the ‘room temperature’ frequently encountered by this synanthropic species, as well as the optimum found by Barghusen et al. (1997). The other temperatures were considered intermediary. Each trial consisted of 4 specimens tested simultaneously at the same temperature. The trials were repeated 3x, for a total sample size of 12 specimens for each temperature treatment.

For each round of trials, specimens were randomized and placed into their temperature treatments. Temperature treatment order within a round was also randomized (Table 1). Each trial consisted of 48 h in a H2200-H MyTemp mini digital incubator (www.benchmarkscientific.com) set to the appropriate temperature. Prior to treatments, 3 wooden dowels (~21.0 cm L × 0.3 cm D) were weighed on a digital scale at 21°C. These dowels were placed into a 17.8 cm L × 11.4 cm W × 15.2 cm H clear plastic box drilled with 1 mm air holes along with a single specimen. Four of the boxes were then placed into the incubator. Temperature and relative humidity in the incubator were monitored and recorded every 12h throughout the duration of the 48h. Since Barghusen et al. (1997) found no effect of humidity on web construction, we did not attempt to equalize humidity levels at the different temperatures.

After a trial ended, survival or mortality of specimens was determined. Constructed webs were wound around the dowels and weighed after returning to 21°C. The web mass was calculated by subtracting the initial mass of the dowel from the final mass of the dowel and web after the test concluded. Spider specimens were preserved in a pre-weighed vial of ethanol. Spider mass was calculated by subtracting the initial mass of the vial from the final mass of the vial with the specimen. Each specimen was treated as an independent observation.

All data analyses were conducted using Excel for Windows with the RealStats add-in (www.real-statistics.com) with non-parametric tests since no resulting data met parametric assumptions. Mean arcsine transformed percent survival of specimens within the temperature treatments were compared via a Kruskal-Wallace test with post-hoc Dunn test. Mean weight of webs of temperature treatments were also compared via a Kruskal-Wallace test with post-hoc Dunn test. Spearman rank-order correlations were calculated between the mass of spiders and the mass of produced webs for each temperature treatment. Acclimation temperature and acclimation humidity were determined by taking the mean of the 5 previous days that the spider had been exposed to before its temperature test. Spearman rank-order correlations were then calculated between the mass of produced web and both ambient acclimation temperature and relative humidity.

### Table 1. Dates of trials and the randomly-assigned order of temperature (°C) treatments for each trial.

<table>
<thead>
<tr>
<th>Trial round</th>
<th>Date begun</th>
<th>Date ended</th>
<th>Temperature treatment order</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>14 May</td>
<td>23 May</td>
<td>50 30 35 40 21</td>
</tr>
<tr>
<td>2</td>
<td>24 May</td>
<td>02 June</td>
<td>35 30 40 21 50</td>
</tr>
<tr>
<td>3</td>
<td>03 June</td>
<td>12 June</td>
<td>40 30 21 35 50</td>
</tr>
</tbody>
</table>

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Table 1. Dates of trials and the randomly-assigned order of temperature (°C) treatments for each trial.

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<td>35 30 40 21 50</td>
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<tr>
<td>3</td>
<td>03 June</td>
<td>12 June</td>
<td>40 30 21 35 50</td>
</tr>
</tbody>
</table>
Results

The 21, 30, and 35°C treatments had the highest survival rate, the 50°C treatment, had the lowest, and the 40°C treatment was intermediate (Fig. 1). The 21, 30, and 35°C treatments had higher web mass than the 40 and 50°C treatments. Correlations between spider mass and web mass were not significant at any temperature (Fig. 2). There was no difference in mean web mass between spiders that died during a 40°C trial and those that lived for the entire trial (Two-sample T-test, df = 10, \( P = 0.19 \)). Ambient room indoor temperature varied from 18–25°C during the 30 days of the experiment, and relative humidity ranged from 43–73% (Fig. 3). Correlations between web mass and both temperature and relative humidity, however, were not significant (Fig. 4,5).

Discussion

This study provides the first preliminary data on 48-h lethal temperature tolerance of adult *P. tepidariorum*. Since 42% of specimens died at 40°C and 100% died at 50°C, the actual upper lethal temperature is somewhere between those values. This result is comparable to that of other tested spider species, ~80% of which had an upper temperature limit of 43–48°C in the laboratory (Mouer and Ericksen 1972, Tolbert 1979, Suter 1981, Pulz 1987). Since temperatures this high are unlikely to be encountered in most environments, their primary importance is in comparison to those of other spiders tested under similar circumstances.

This study also provides evidence that *P. tepidariorum* has a fairly wide optimal temperature range for web production. Although they claimed 20°C as the optimal temperature for prey catch, Barghusen et al.’s (1997) study of *P. tepidariorum* demonstrated no statistical difference in web mass between 15–30°C. Similarly, we observed no difference between 21–35°C, suggesting an ability of this cosmopolitan species to effectively construct webs over a temperature range of 15–35°C.
Our results also suggest an inability of *P. tepidariorum* to produce webs beginning at ~35–40°C. While the web mass of spiders in the 35°C trials approximated that of spiders at lower temperatures, the web mass of spiders at 40°C was the same as that of spiders at 50°C, all of whom died and spun no web (Fig. 1). Moreover, there was no difference in the web mass of spiders that survived to the end of the 40°C trial and those that died at this temperature. This result argues strongly for sublethal web mass decrease, because if spiders were spinning less web due to mortality, then there would have been a difference in web mass between spiders who survived the trials and those that did not.

This result demonstrated the sub-lethal effect of high temperature, as a prolonged exposure to ~40°C in nature might ultimately prove fatal to the species due to its inability to produce a web, even without direct temperature mortality. Small decreases in web mass can have significant decreases in capture efficacy, especially when coupled with the increased metabolic activity and mechanical power of prey insects at warmer temperatures (Hesselberg and Vollrath 2006). Daytime temperatures near 40°C can realistically occur in most temperate environments, albeit for only a portion of the day, and are likely to become more common in the future (e.g., Dai et al. 2016). Since *P. tepidariorum* is a cosmopolitan species with a fairly wide optimal temperature range, it is likely that other spider species may experience sub-lethal high temperature effects at lower temperatures.

The size of our spider specimens appeared to have no effect on the mass of their webs. The relationship between spider mass and web mass is not well understood. Blackledge and Zevenbergen (2007) found that recently fed, and thus larger, specimens of *Latrodectus hesperus* Chamberlin & Ivie constructed heavier webs than did fasted specimens. This result was in contrast to studies of orb weaving spiders (Araneidae), which built smaller webs as they caught more food (Sherman 1994; Venner et al. 2000, 2006), or because they allocated more energy to foraging effort instead of web construction (Venner et al. 2003). Since *P. tepidariorum* is more closely related to *L. hesperus*, and constructs a similar type of web, we suspect it should have a similar relationship between spider mass and web mass. Because we were consistent in our feeding, any effects of hunger should have been similar between specimens and, thus, differences in body mass should have been
due primarily to inherent size difference. Such size differences appeared to have no effect on web mass.

One potential source of error in our study was the fluctuating temperature and associated relative humidity during our acclimation period. The temperature at which an organism is acclimated prior to testing has been shown in numerous studies to have a significant effect on its laboratory-determined thermal tolerance (e.g., Lutterschmidt and Hutchison 1997). We, however, found no correlation between web mass and either temperature or relative humidity based on the mean conditions during the 5 d before a trial, despite such conditions varying by ~5°C. One possible reason for this lack of acclimation is that in intentional laboratory acclimation trials, temperatures are held at a constant temperature for a prescribed period, whereas our temperatures fluctuated based on local weather. Such fluctuations are typical of natural temperate environments, and may lead to an increased temperature tolerance range of specimens exposed to them relative to those exposed to constant temperature (Li and Jackson 1996). Thus, the varying temperatures in our study probably did not strongly acclimate a specimen to a particular temperature, and any minor acclimation effects were insignificant.

Further research is needed to translate our laboratory results to natural environments. Specifically, longer trials at temperatures with differences <5°C between them will help determine specific lethal and sub-lethal threshold temperatures. Because warm temperatures may ultimately harm the spider through starvation, specific experiments on the length of time specimens can live without food at high temperatures are also important, since a long ability to survive without eating would offset our observed sub-lethal effects. Lastly, doing the above experiments with more stenothermic species may suggest how warming environments may affect the important ecological services provided by spiders.

Acknowledgments

We thank Carmen, Garth, and Mikayla Brown for assisting us with laboratory equipment, and Angie Pytel for editing an earlier version of the manuscript. Research costs were supported by the Hillsdale College biology department. This paper won the best student presentation award at the 64th Annual Meeting of the Michigan Entomological Society. It is paper #21 of the G.H. Gordon BioStation Research Series.

Literature Cited


Leaf Mining Insects and Their Parasitoids in the Old-Growth Forest of the Huron Mountains

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Abstract

Leaf mining insects in an old-growth forest along the south central shore of Lake Superior in Michigan are documented. We present the results of a 13-year survey of leaf mining species, larval hosts, seasonal occurrence, and parasitoids, as well as report biological observations. Representative larvae, mines, adults, and parasitoids were preserved. Among the larval host associations, 15 are reported as new. Additionally, 42 parasitoid taxa were identified resulting in six first reports from the New World and 32 new host associations. Two undescribed species (Gelechiidae and Figitidae) discovered through this research were described in earlier publications.

Keywords: biodiversity, Chalcidoidea, Coleoptera, Diptera, Hymenoptera, Ichneumonoidea, Lepidoptera, Nearctic

The leafmining guild is an important ecological component of forest biodiversity. Leaf miners occur primarily among Diptera and Lepidoptera with only a few species among the Coleoptera and Hymenoptera (Needham et al. 1928). In their larval form, leaf miners feed inside one or more leaves for all or at least a portion of that stage. Their mining patterns can vary considerably among species, from a narrow sinuous trail to a large blotch encompassing the entire leaf. Mines may occur on the upper-, the lower-, or both leaf surfaces as the larvae grow. While a mine provides its inhabitant with a protective cover, it can also function as its prison. For example, miners have little escape from a variety of parasitic Hymenoptera that attack them. In spite of their variously shaped mines, their small size and unobtrusive feeding habits allow these insects to be easily overlooked as a subject of study in forest ecosystems.

One forest type in northern Michigan, is the Hemlock-White Pine-Northern Hardwoods which once encompassed over 15 million hectares (Frelich and Reich 1996, Dickman and Leefers 2003). This forest type extended “from northern Minnesota and extreme southeastern Manitoba through the upper Great Lakes region and eastward across southern Canada and New England” (Braun 1950). A remnant of old-growth Hemlock-White Pine-Northern Hardwoods forest occurs within a private land holding called The Huron Mountain Club (HMC) located in the upper peninsula of Michigan (Fig. 1). Many of the components of this old-growth forest have been studied (see: www.hmwf.org/archives/reports/), however, not leaf miners.

The landscape of the HMC is composed of Precambrian metamorphic bedrock hills (Dorr and Eschman 1970, Simpson et al. 1990). “The metamorphic rock, much of it over 2.5 billion years old, is a part of the Canadian Shield and in the Huron Mountains has been differentially eroded to produce the current bedrock topography” (Simpson et al. 1990). Its physiography is one of hills in sharp relief, 10 intermontane lakes, and a diverse array of old-growth forests. Soils vary from nearly absent in mountain crevices to deep in the low flat lands and sandy along its beaches (Braun 1950). Cool temperatures and a low evaporation rate characterize the climate of HMC. Except for the first 3–5 km along Lake Superior, the lake does not significantly moderate land temperatures since prevailing winds are southwesterly (Denton and Barnes 1988).

Simpson et al. (1990) studied the landscape of HMC and divided the area into 29
cover types with 22 upland types covering 94% of the area. Of those, the two larger cover types are hemlock-northern hardwood (49%) and lichen-juniper, pine-oak, and pine-hemlock hardwoods (30%). Other upland types include pine, birch-aspen, and wet site conifer and conifer-hardwood. The remaining wetland cover types include open and forested land. These cover types, with nine lakes and several streams, contribute to its diversity of life supporting leaf mining insects, their hosts, and parasitoids.

The main objective of our 13-year survey was to document leaf miner habitats: 1) advice from knowledgeable individuals who frequently guided the first author (RJP) to sites, 2) literature descriptions (Etter et al. 1960, Wells and Thompson 1976, Rydholm 1989, Simpson et al. 1990, Manierre 1999), and 3) exploration of areas via 2-track roads and walking trails. Habitats surveyed included fen, bog, beach, old field, stream flood plain, beech/maple forest, deciduous forest undergrowth, and some areas of disturbed ground along main roads. The sites were sampled for 93 cumulative sampling days from 2000–2012. For many sites sampling was conducted multiple times during various seasons and over multiple years. This provided a greater chance of collecting species that were rare or had fluctuating population numbers. Previous survey experience in upper Michigan by the first

Materials and Methods

Three methods were used to locate leaf miner habitats: 1) advice from knowledgeable individuals who frequently guided the first author (RJP) to sites, 2) literature descriptions (Etter et al. 1960, Wells and Thompson 1976, Rydholm 1989, Simpson et al. 1990, Manierre 1999), and 3) exploration of areas via 2-track roads and walking trails. Habitats surveyed included fen, bog, beach, old field, stream flood plain, beech/maple forest, deciduous forest undergrowth, and some areas of disturbed ground along main roads. The sites were sampled for 93 cumulative sampling days from 2000–2012. For many sites sampling was conducted multiple times during various seasons and over multiple years. This provided a greater chance of collecting species that were rare or had fluctuating population numbers. Previous survey experience in upper Michigan by the first
The author found a flush of miners early in the growing season, a reduction in July and August, and then the highest abundance in late summer and autumn. These observations guided the timing of the current surveys (Table 1). Rearing larvae from host foliage was the exclusive method for obtaining leaf miner data, including larval hosts, mines, immaculate adults, seasonal occurrence, and parasitoids. Active miners were recovered by visually searching leaves between 0–2 m above ground. Leaves containing similar mines from the same host species and location, termed a “lot”, were placed in a single quart-size freezer Ziploc® bag with a field identifying label. Each mine of taxa known to pupate outside its mine, such as Agromyzidae, Nepticulidae, and Tenthredinidae, were placed in separate bags when collected. Upon returning from the field each lot was assigned a unique sequential lot number, recorded on a separate rearing notes form. Each larva within each lot was also given a unique number preceded by its associated lot number. Issuing parasitoids were likewise numbered with their associated host’s number. All relevant data for each specimen were recorded on the rearing notes forms. Each leaf was placed in a separate bag with its assigned specimen number. All bags were held in translucent plastic gallon boxes fitted with air-tight lids. Mines were observed daily and data recorded on mine shape, feeding patterns, and larval habits.

When miners pupated in their mines, the mined leaves were placed in vials for adults to issue. Vials were held in sealed gallon plastic boxes with slightly moistened paper toweling and checked daily for adult issue. Immatures recovered in the fall were wintered in bags or vials inside gallon plastic boxes in an unheated home garage. Wintering mines were examined weekly to confirm presence of adequate moisture. Boxes were brought indoors by mid-February to force adult emergence, which occurred between March and May for most overwintering individuals. Vacated mines were pressed, then preserved in glassine envelops with a label containing lot number, host plant, and the mining species. Adult miners were double mounted (Landry and Landry 1994). Issuing parasitoids were killed, mounted on points, card-mounted (Noyes 1982), or preserved in 80% ETOH.

While larvae fed, representative mines were photographed (Priest 2007), and some were preserved as described by Stehr (1987). All microscopical observations were made using a Leitz Wetzlar wide-field stereomicroscope with eyepiece reticle. Images of adult miners were photographed and assembled with a Visionary Digital Passport II system (Palmyra, VA) using a Canon EOS 5D Mark II, 58.0 mm Canon Macro photo lens, Canon Speedlite transmitter ST-E2, two Canon Speedlite 430EX II flashes and a Stack Shot (Cognisys, Inc., Kingsley, MI) controlled by Zerene Stacker 1.04 (Zerene Systems LLC, Richland, WA). Montage images were assembled using Helicon Focus Mac Pro 4.2.8 (Helicon Soft, Kharkov, Ukraine).

Adult miners were identified primarily by the first author using the following literature: COLEOPTERA: Buprestidae: Wellso et al.: TGLE Vol. 52 Nos. 3 & 4 Full Issue
al. (1976), Bright (1987), Downie and Arnett, Jr. (1996), comparison with specimens at the A. J. Cook Arthropod Research Collection, Michigan State University (MSUC); Chrysomelidae: Parry (1974); Staines (2006); names used according to Clark et al. (2004). DIPTERA: Agronyzidae: mines of *Liriomyza robini*ae Valley tentatively identified with Weaver and Dorsey (1967) and Valley (1982); *Phytomyza plantaginis* Robineau-Desvoidy tentatively identified with Spencer (1973). HYMENOPTERA: Tentredinidae: *Betula*-feeding species were identified using Digweed at al. (2009); other species identified using Smith (1971, 1988) and Hoebeck and Wheeler, Jr. (2005). LEP-IDOPTERA: Argyresthiidae: identified with Freeman (1972); Bucculatricidae: identified with Braun (1965); Eriocraniidae: tentative-ly identified with Davis (1978, 1987) and Stehr and Martina (1987); Gracillariidae: Dietz (1907), Braun (1908), Forbes (1923), Weaver and Dorsey (1967), and Maier and Davis (1989); Nepticulidae: Braun (1917) and Wilkinson and Scoble (1979); Tischeridae: Braun (1972). Adult Lepidoptera were also compared with specimens at the USNM. Specimens from rearings which produced no adults were included and cited as “probably” (prob.) if the larva and mine were in concor-dance with known geography, seasonality, host plant, mine placement and shape, frass arrangement, larval color, and pupation site of the cited species. Plants were identified by RJP using Cobb (1963), Voss (1972, 1985, 1996), Wells and Thompson (1976), and Voss and Reznicek (2012).

The second author (RRK) identified all Braconidae using Leica Wild M10 and Leica M205 A stereomicroscopes with 10X or 25X oculars. All specimens were determined to genus using the relevant keys to genera in Wharton et al. (1997). Specimens were identified to species, when possible, using relevant keys listed in Yu et al. (2012). All species identifications were corroborated through comparison with specimens in the USNM identified previously by braconid systematists, Yu et al. (2012) was used for *Brac-onidae and Ichneumonidae* classification as a standard, but use of the classification herein should not imply that the authors agree with the placement or status of all taxa.

The third author (MWG) identified all Chalcidoidea using a Nikon SMV-1500 stereomicroscope with 10X oculars. All specimens were determined to genus by sight identification or using Gibson et al. (1997). Specimens were identified to species, when possible, using relevant keys listed in Noyes (2018). All species identifications were corroborated through comparison with authoritatively identified specimens in the Smithsonian Institution National Museum of Natural History (USNM).

Most voucher specimens of adult miners, larvae, mines, and rearing note were deposited in MSUC. The remaining voucher specimens were deposited in the collections at the institutions of the experts providing those identifications (see Acknowledgments). Some host plants were deposited at the University of Michigan and Michigan State University herbaria. Though most plants were not vouchered at a herbarium, leaves with mines were pressed and placed in MSUC. Most preserved leaves and many of the illustrated mines show a sufficient amount of the host leaf to make plant identification possible. Lists of parasitoids reported from the hosts treated herein were obtained primarily from Noyes (2018) and Yu et al. (2012). Other sources are identified with the specific miner species’ parasitoids discussed.

### Results and Discussion

A total of 221 lots were reared resulting in 63 leaf miner taxa and 42 parasitoid taxa identified (Tables 2 and 3). Thirteen larval-host plant species associations and three larval-host plant genera associations are newly reported. Twenty leaf miners are reported as new to Michigan. Scrobipalpula *manierreorum* Priest (Gelechiidae) and *Ban-acuniculus strykeri* Buffington (Figitidae) were newly discovered during this survey and described prior to this paper. Additionally, we report the first rearing of *Chriosia spinosissima* (Malloch) (Anthomyiidae) from larva to adult. We also document miners from 38 plant taxa in 13 plant families. Forty-two parasitoid taxa in six families of Hymenoptera were identified with six New World records.

The plant survey by Wells and Thompson (1976) included approximately 90 square miles of the Huron Mountain range and identified an extant 781 species. In the current survey, mines were reared from only 41 plant taxa. The first author has observed, while surveying leaf miners throughout Michigan, that the farther north surveys occur, numbers of both species abundance and richness decline. As a result, longer periods are required in northern regions compared to southern regions of Michigan to obtain even modest results. The presence of vacated and unfamiliar mines during this survey suggests many additional mining species await discovery in this remarkable old growth land holding and throughout Michigan. We report data from each reared species below.
COLEOPTERA

Buprestidae

Brachys aerosus (Melsheimer) (Fig. 2, 3). Specimen data: Breakfast Roll Mountain 46°51.051N 87°49.601W, 2 larvae 25 Sep 2000, 3 adults after wintering, Lot 1088; same location, 4 larvae 26 Sep 2001, 2 adults after wintering, Lot 1261; River Styx 46°50.627N 87°51.300W, 10 larvae 27 Sep 2001, 3 adults after wintering, Lot 1247; same site, 2 larvae 25 Sep 2008, 2 adults after wintering, Lot 1905; Lily Lake 46°50.892N 87°49.783W, 4 larvae 26 Sep 2002, 3 adults after wintering, Lot 1361; same site, 2 larvae 16 Sep 2005, 1 adult after wintering, Lot 1658. Distribution: CANADA: AB, BC, MB, NB, ON, QC, SK, U.S.A.: AL, AR, AZ, CT, FL, GA, IA, IL, IN, MA, MD, ME, MI, MO, MT, NC, ND, NH, NJ, NY, OH, OK, PA, RI, SC, SD, TX, VT, VA, WA, WI, WV (Nelson et al. 2008). Larval hosts: Haricots, usually oak (Wells and Thompson 1976); Acer sp., Castanea sp., Cornus sp., Fagus sp., Hamamelis virginiana L., Populus tremuloides Michx., Quercus sp., Tilia americana L., Ulmus sp. (Bright 1987); Quercus rubra L. var. ambigua (Gary) Fern. (Nelson et al. 2008). Except for Nelson et al. (2008), the previous references do not specify hosts as either of larvae or adults. Quercus rubra (identified in this survey). Parasitoids: Eulophidae: Closterocerus cinctipennis Ashmead (Noyes 2018). Remarks: Only one mine per leaf, and a single larva in each mine was observed. "maple reproduction layer." There is only one mine per leaf and one larva per mine. The full depth mine is a wide track; because the winding tracks are frequently contiguous, it may appear as an irregular blotch. The later feeding area is light green though most of the mine is tan colored. Short black string frass forms irregular trails. Some larvae were observed exiting the lower surface of their mine prior to wintering, but these specimens died. Larvae usually winter in their mines with pupation occurring after winter and adults issuing within two weeks of pupation. There is only one generation per year at HMC.

Chrysolinaeidae

Dibolia borealis Chevrolet (Fig. 6, 7). Specimen data: Ken Cass drive near Lily Pond 46°50.892N 87°49.783W, 9 larvae 14 Jul 2003, 0 adults, Lot 1414; near Lower Falls 46°48.720N 87°48.772W, 40+ larvae 08 Jun 2006, 0 adults, Lot 1701. Distribution: CANADA: AL, MB, NB, NS, ON, PE, QC, U.S.A.: AL, AR, CT, DC, FL, IA, IL, IN, KS, LA, ME, MD, MA, MI, MS, MO, MT, NH, NJ, NY, NC, OK, RI, SC, TN, TX, VT, VA, WV, WI (Parry 1974). Larval hosts: Plantago major L., P. lanceolata L., P. rugelii Decne. (Clark et al. 2004); P. major (identified in this survey). Parasitoids: Eulophidae: Chrysocharis nitetis (Walker) (Noyes 2018). Remarks: Several mines were found per leaf but only one larva per mine. The mine is full depth, meandering and serpentine, with a black central frass trail. Two rearings of feeding larvae did not produce adults though adults were collected on mined leaves. The
### Table 2. Reared leaf mining insects, hosts, and parasitoids. Acronyms are: FH = first reported larval host; NG = newly reported host genus; NO = newly reported Michigan occurrence; NS = newly reported host species. Months are divided into thirds: E (early) = 1st-10th; M (mid) = 11th-20th, L (late) = 21st to month end. Parasitoid numbers refer to Table 3.

<table>
<thead>
<tr>
<th>Leaf Miner</th>
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<th>Recovery Period</th>
<th>Emergence Period</th>
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<tr>
<td>Buprestidae</td>
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<td>Brachys aerosus</td>
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<td>M-L Sep</td>
<td>wintered</td>
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<td>Brachys aeruginosus</td>
<td>Acer saccharum</td>
<td>M-L Sep</td>
<td>wintered</td>
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<tr>
<td>Chrysomelidae</td>
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<tr>
<td>Dibolia borealis</td>
<td>Plantago major</td>
<td>E-Jun-M Jul</td>
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<tr>
<td>Sumitrosis inaequalis</td>
<td>Eurybia macrophylla</td>
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<td>DIPTERA</td>
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Continued on next page
Table 2. Continued.

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<td>Quercus rubra</td>
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Table 3. Hymenopteran parasitoids reared from leaf miners collected at The Huron Mountain Club in upper Michigan. Emergence periods: E (early) = 1st-10th of month; M (mid) = 11th-20th of month; L (late) = 21st-end of month. Recv. = Recovery period. Emerg. = Emergence period.

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<td>Ceradontha sp. poss. morosa</td>
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<td>Liriomyza lathyri*</td>
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<td>E Sep</td>
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<td>Liriomyza lathyri*</td>
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<td>Colastes sp. 1</td>
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<td>wintered</td>
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<tr>
<td>Dacnusa (Papysnema) sp. poss. discolor (Förster)**</td>
<td>Phytomyza loewii</td>
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<td>Dapsilarthra (Dapsilarthra) rufiventris (Nees)**</td>
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<td>E Jun</td>
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<td>E Aug</td>
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<td>M Aug</td>
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<td>Phyllonorycter martiella*</td>
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<td>Chrysocharis ainsliei Crawford</td>
<td>Liriomyza frickii</td>
<td>M Jun</td>
<td>L Jun</td>
</tr>
<tr>
<td>Chrysocharis beckeri Yoshimoto</td>
<td>Cerodontha sp. poss. morosa*</td>
<td>L Jun</td>
<td>M Jul</td>
</tr>
<tr>
<td>Chrysocharis crassiscapus (Thompson)</td>
<td>Amauromyza flavifrons</td>
<td>L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Chrysocharis giraulti Yoshimoto</td>
<td>Amauromyza flavifrons</td>
<td>L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Chrysocharis occidentalis (Girault)</td>
<td>Leucanthisa directa</td>
<td>M-L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Chrysocharis occidentalis (Girault)</td>
<td>Amauromyza flavifrons</td>
<td>L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Chrysocharis sp. (male)</td>
<td>Amauromyza flavifrons</td>
<td>L Jun</td>
<td>M Jul</td>
</tr>
<tr>
<td>Chrysocharis sp. (female)</td>
<td>Phytomyza loewii*</td>
<td>L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Chrysocharis sp.</td>
<td>Leucanthisa directa*</td>
<td>L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Closterocerus trifasciatus Westwood</td>
<td>Phyllocnistis populatella</td>
<td>M Aug</td>
<td>L Aug</td>
</tr>
<tr>
<td>Closterocerus trifasciatus Westwood</td>
<td>Phyllocnistis populatella</td>
<td>M Aug</td>
<td>L Aug</td>
</tr>
<tr>
<td>Clorois sciericornis (Nees)</td>
<td>Macroacaus robinia*</td>
<td>L Sep</td>
<td>wintered</td>
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<tr>
<td>Clorois sciericornis (Nees)</td>
<td>Paroxysm conspicuella*</td>
<td>L Sep</td>
<td>wintered</td>
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<tr>
<td>Pnigalio maculipes (Crawford)</td>
<td>Leucanthisa directa*</td>
<td>L Sep</td>
<td>wintered</td>
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<tr>
<td>Pnigalio maculipes (Crawford)</td>
<td>Paroxysm conspicuella*</td>
<td>L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Pnigalio maculipes (Crawford)</td>
<td>Macroacaus robinia*</td>
<td>M Aug</td>
<td>L Aug</td>
</tr>
<tr>
<td>Pnigalio sp.</td>
<td>Macroacaus robinia*</td>
<td>M Aug</td>
<td>L Aug</td>
</tr>
<tr>
<td>Pnigalio sp.</td>
<td>Leucanthisa directa*</td>
<td>L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Pnigalio sp.</td>
<td>Macroacaus robinia*</td>
<td>M Aug</td>
<td>L Aug</td>
</tr>
<tr>
<td>Pnigalio sp.</td>
<td>Paroxysm conspicuella*</td>
<td>L Sep</td>
<td>wintered</td>
</tr>
<tr>
<td>Pnigalio sp.</td>
<td>Phyllocnistis populatella</td>
<td>M Aug</td>
<td>L Aug</td>
</tr>
<tr>
<td>Tetrastichos sp.</td>
<td>Macroacaus robinia*</td>
<td>M Aug</td>
<td>L Aug</td>
</tr>
</tbody>
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*First report from this host. See text. **New World record.
Figures 2–9. Fig. 2. *Brachys aerosus*: adult. Fig. 3. *Brachys aerosus*: nearly completed mine with larva. Fig. 4. *Brachys aeruginosus*: adult. Fig. 5. *Brachys aeruginosus*: nearly completed mine with larva. Fig. 6. *Dibolia borealis*: adult. Fig. 7. *Dibolia borealis*: several mines, each with a single larva. Fig. 8. *Sumitrosis inaequalis*: adult. Fig. 9. *Sumitrosis inaequalis*: completed and vacated mine.
larva is dark yellow with a dark brown head and prothorax. It exits its mine prior to pupating. These observations agree with those of Reed (1927). Adults were recovered on host leaves during early June only.


DIPTERA

Agromyzidae

Agromyza canadensis Malloch (Fig. 10). Specimen data: Stone House 46°50.627N 87°51.300W, 10 larvae 30 Jun 2000, 1♂ 30 Jun 2000, Lot 1024. Distribution: CANADA: AL, ON, YT; U.S.A.: CA (Frick 1959, Sehgal 1971, Spencer 1981). Larval hosts: Mertensia paniculata (Nixon), P. norvegica (Walker); E. divaricata (Engelm.) Nesom, Zexmenia sp. (Staines 2013). Parasitoids: Opius sp., Braconidae: E. cyclogaster Förster (identified in this survey). Remarks: There are two to nine mines per leaf and one to three larvae per mine. The mine is an upper surface wide meandering track formed with the initial portion darkened. Frass is composed of black irregular particles in a distinct central trail. The larva exits the upper leaf surface prior to pupating. Prior to this research E. cyclogaster was known only from the Palearctic Region, ranging in the west from the United Kingdom east to Azerbaijan but has also been reported from Primorsky Krai, Russia (Yu et al. 2012). The specimens in this research are the first records of this species for the New World and the first braconid reported from A. canadensis.

Agromyza isolata Malloch (Fig. 11). Specimen data: Main and Flat Rock roads 46°53.651N 87°55.171W, 10 larvae 18 Sep 2007, 5 adults 07–08 Oct 2007 and 1♂ after winter, Lot 1837. Distribution: U.S.A.: CA, CO, KS, MN, PA, WA (Spencer and Steyskal 1986). Larval hosts: Populus spp., Salix spp. (Spencer and Steyskal 1986). Populus grandidentata Michx. (identified in this survey). Parasitoids: Braconidae: C. isolatae Fischer (Yu et al. 2012). Braconidae: Exotela cyclogaster Förster (identified in this survey). Remarks: There are two to nine mines per leaf and one to three larvae per mine. The mine is a wide surface meandering track formed with the initial portion darkened. Frass is composed of black irregular particles in a distinct central trail. The larva exits the upper leaf surface prior to pupating. Prior to the research E. cyclogaster was known only from the Palearctic Region, ranging in the west from the United Kingdom east to Azerbaijan and in the east from Sakhalin Oblast, Russia to the Korean Peninsula (Yu et al. 2012). The specimens in this research are the first records of this species for the New World and as a parasitoid of A. isolata.

Agromyza sp. prob. potentillae (Kaltenbach) (Fig. 12). Specimen data: Loop Rd. 46°50.627N 87°51.300W, 6 larvae, 20 Sep 2007 0 adults, Lot 1845. Distribution: CANADA: Widespread; U.S.A.: CA, CO, UT (Spencer and Steyskal 1986); CANADA: BC, ON, QC (as A. spiraeae) (Spencer 1969). Larval hosts: Potentilla gracilis Douglas ex Hook. (Spencer and Steyskal 1986), Potentilla norvegica L. (identified in this survey). Parasitoids: Eulophidae: P. semiomus (Walker); Pteromalidae: Stictomischus groschkei Delucchi (Noyes 2018). Braconidae: Chorebus (Strophocera) bres (Nixon), Chorebus (Strophocera) cedrata (Nixon), Chorebus (Strophocera) deione (Nixon), Chorebus (Strophocera) eros (Nixon), Dacnusa (Pachysema) evadne Nixon,
Dacnusa (Pachysema) laeta (Nixon), Dacnusa (Dacnusa) maculipes Thompson, Dapsilartha (Heterolexis) balteata (Thompson), Dapsilartha (Heterolexis) okazaki Takada and Imura, Dapsilartha (Dapsilartha) rufiventris (Nees), Exotela nowakowskii Griffiths, Opium (Opium) agromyzicola Fischer, Opium (Nosopoea) cingulatus Wesmael, Opium (Nosopoea) maculipes Wesmael, Opium (Opium) pallipes Wesmael, Opium (Agnopius) similis Szépligeti, Opium (Allophlebus) singularis Wesmael (Yu et al. 2012), Braconidae: Dapsilartha sp. indet. (identified in this survey). **Remarks:** There is only one mine per leaflet and one larva per mine. The mine is located on the upper surface, initially serpentine but expanding into an elongate blotch. Frass is black and scattered. Prior to pupating the larva exits the upper leaflet surface. The only other known North American agromyzid blotch miner of Potentilla is Agromyza sulfuriceps Strobl. Its frass is irregular black particles scattered through the leaf. The larva crosses the main vein while feeding and exits the upper leaflet surface prior to pupating. The fully developed last instar larva is golden yellow. The Holarctic Chrysocharis crassiscapus is reported from A. flavifrons for the first time. It was previously reported from Amauromyza sp. by Hansson (1987).

Cerodontha sp. poss. morosa (Meigen) (Fig. 15). Specimen data: Mountain Stream Falls 46°52.188N 87°53.685W, 1 larva, 37 puparia 2 Jun 2000, 1 adult 07 Jul and 14 Jul 2000, Lot 1022. Distribution: U.S.A.: CA, MT (Spencer and Steyskal 1986). Larval hosts: Carex (Spencer and Steyskal 1986). Carex sp. (identified in this survey). Parasitoids: Braconidae: Chorebus (Stiphrocerca) pelion (Nixon), Chorebus (Chorebus) siniffa (Nixon), Opium (Agnopius) similis (Yu et al. 2012). Eulophidae: Chrysocharis beckeri Yoshimoto, Pedioibius albipes (Provancher); Pteromalidae: Halictoptera sp. (identified in this survey); Braconidae: Chorebus (Stiphrocerca) sp. poss. pelion (identified in this survey). **Remarks:** There are one to four mines per leaf with one larva per mine. The completed upper surface mine is a longitudinal track, crossing larger veins only when the larva is nearly fully developed. Frass is not apparent. The larva pupates within its mine. Host, mine, shape, location, and pupation site are consistent with the habits described for C. morosa in Spencer and Steyskal (1986). The species could not be identified, “Apex of distiphallus absent,” Boucher, personal communication. Chorebus pelion is currently known only from the Palearctic Region. It has been reported primarily from Sweden south to Germany and east to Ukraine but has also been reported from Kamchatka Krai, Russia (Yu et al. 2012). If verified the specimen in this research would be the first record of C. pelion for the New World. Chrysocharis beckeri, P. albipes, and Halictoptera sp. are all new host records for C. morosa.

Figures 10–17. Fig. 10. *Agromyza canadensis*: vacated mine showing arced feeding pattern. Fig. 11. *Agromyza isolata*: 2 coalesced and vacated mines. Fig. 12. *Agromyza sp. prob. potentillae*: 2 leaflets, each with a single mine, 1 with an active larva. Fig. 13. *Amauromyza flavifrons*: adult. Fig. 14. *Amauromyza flavifrons*: completed and vacated mine. Fig. 15. *Cerodontha sp. poss. morosa*: completed and vacated mine. Fig. 16. *Liriomyza brassicae*: adult. Fig. 17. *Liriomyza brassicae*: nearly completed mine with larva at mine widening.
Hooker (identified in this survey). **Parasitoids: Eulophidae: Chrysocharis sp.**, Chrysocharis vonones (Walker), Closterocerus mirabilis Edwards and La Salle, Diaulolinopsis calliechroma Crawford, Diglyphus sp. Walker, *Meruacea liriomyzae* (Boucek), *Pnigalo* sp. **Pteromalidae: Pachyneurosp**, Trigonogaster parasitica Girault (Noyes 2018); **Braconidae: Opisus (Pendopius) volaticus* Fischer, *Phaedrotoma scabriventris* (Nixon) (Yu et al. 2012). **Eulophidae: Chrysocharis ainslei** Crawford (identified in this survey). **Remarks:** The mine is located on both leaf surfaces though more apparent from the upper side. It is linear gradually widening and frequently traversing much of the leaf length. Black frass is placed in an intermittent trail along one side of the mine. The larva exits the upper leaf surface prior to pupating. *Chrysocharis ainslei* is reported for the first time from *L. brassicae*. Its known dipteran host range includes six genera of Agromyzidae, including 10 species of *Liriomyza*. It has also been recorded from four families of microlepidoptera, one species each (Noyes 2018).

**Liriomyza eupatorii** (Kalthenbach (Fig. 18, 19). **Specimen data:** Lower Dam 46°48.720N 87°48.772W, 3 larvae, 1 puparium 13 Jun 2001, 1♂ 24 Jun 2001, Lot 1129. **Distribution:** EUROPE, CANADA: AB; U.S.A.: CA, GA, MS, NC, SC, TN, VA, WA (Sehgal 1971, Spencer and Steyskal 1986). **Larval hosts:** Symphyttrichum (Aster) chilensis (Noes) G. L. Nesom, Eupatorium, Helianthus, Lampsana (misspelling, possibly *Lapsana*, an Asteraceae), Solidago sp. (Sehgal 1971, Spencer and Steyskal 1986). Solidago sp. (identified in this survey). **Parasitoids: Eulophidae: Chrysocharis liriomyzae** Delucchini, Chrysocharis pentheus, *Pnigalo soemius*; **Pteromalidae: Miscogaster** sp., Miscogaster maculata Walker, Sphaeripalpus fuscipes (Walker) (Noyes 2018); **Braconidae: Atorum vietus** (Ha-liday), *Opisus (Allophlebus) singularis* (Yu et al. 2012). **Remarks:** Four or more upper surface mines may occur per leaf with a single larva in each mine. Spencer and Steyskal (1986) indicated that the mine in *Solidago* from Mississippi begins as a spiral. Mines in this rearing did not exhibit that feature though a half circle was formed initially. Frass particles are placed in a central broken line. The larva is yellow and exits its mine from either the upper or lower surface prior to pupating. **Liriomyza fricki** Spencer (Fig. 20, 21). **Specimen data:** Salmon Trout Bay 46°51.870N 87°46.435W, 4 larvae 11 Jun 2001, 1♂ 22 Jun 2001, Lot 1126; Conway Bay 46°53.101N 87°48.705W, 4 larvae 23 Sep 2002, 1♂ 06 Oct 2002 and after wintering, Lot 1348. **Distribution:** CANADA: AB; U.S.A.: CA, WA, WI (Sehgal 1971, Spencer and Steyskal 1986). **Larval hosts:** *Lathyrus ochroleucus* Hook., *Medicago, Melilotus* [sic] (probably *Melilotus*), *Trifolium repens* L., *Vicia americana* Muhl., *Vigna* (Sehgal 1971, Spencer and Steyskal 1986). *Lathyrus japonicus* Wild, (identified in this survey). **Parasitoids: Eulophidae: Chrysocharis crassiscapus*, *Chrysocharis oscinidis* (Noyes 2018). **Eulophidae: Chrysocharis ainslei;** **Braconidae: Opisus sp.** 1 (identified in this survey). **Remarks:** The rather short mine is placed on the upper surface with one to two mines per leaf and a single larva per mine. It is linear with a thin central broken green-black frass trail. Mines from these rearings agree with the description in Spencer and Steyskal (1986). The larva is bright yellow upon exiting the upper leaf surface prior to pupating. The pupa is a dull orange. From these two rearings there are clearly at least two generations per year at HMC. The species of *Opisus* reared in this survey is the first braconid reported from *L. fricki*. *Chrysocharis ainslei* is reported for the first time from *L. fricki* (see notes under Discussion of *L. brassicae*).

**Liriomyza lathyri** Sehgal (Fig. 22, 23). **Specimen data:** Conway Bay 46°53.101N 87°48.705W, 2 larvae 24 Sep 2002, 1♂ after wintering, Lot 1349; Gillet Landing 46°51.870N 87°46.435W, 7 larvae 16 Aug 2004, 0 adults, Lot 1508; same site, 4 larvae 16 Sep 2006, 0 adults, Lot 1755; same site, 3 larvae 27 Aug 2009, 1♂, 1♀ after winter, Lot 1939. **Distribution:** CANADA: AB; U.S.A.: AB (Spencer and Steyskal 1986). **Larval hosts:** *Lathyrus ochroleucus* (Sehgal 1971); *Lathyrus sp.* (Spencer and Steyskal 1986). *L. japonicus* (identified in this survey). **Parasitoids: Eulophidae: Chrysocharis crassiscapus** (Noyes 2018). **Braconidae: Chorebus** sp. 3 (identified in this survey). **Fi-gitidae: Bananuciculus stykeri** (holotype). *Bananuciculus stykeri* data were originally published in Buffington (2010). **Remarks:** The upper surface blotch mine is initially linear. One or two mines occur per leaflet with one to four larvae per mine. Frass appears as indistinct scattered green/black spotting. The larva exits the upper mine surface prior to pupating. These observations agree with Sehgal’s (1971) notes in his original species description. The species of *Chorebus* reared in this survey is the first braconid reported from *L. lathyri*.

**Liriomyza sp. prob. robiniae** Valley (Fig. 24). **Specimen data:** Ives Road 46°50.475N 87°50.581W, 10 larvae 10 Jun 2004, 0 adults, Lot 1486. **Distribution:** U.S.A.: NY, PA, VA, WV (Valley 1982). **Larval hosts:** Robinia pseudoacacia L. (Valley 1982). Robinia pseudoacacia (identified in this survey). **Parasitoids:** None reported.
Figures 18–25. Fig. 18. *Liriomyza eupatori*ii: adult. Fig. 19. *Liriomyza eupatori*ii: a completed and vacated mine on each side of main vein. Arc exit cut visible at mine terminus. Fig. 20. *Liriomyza fricki*: adult. Fig. 21. *Liriomyza fricki*: completed and vacated mine. Fig. 22. *Liriomyza lathyri*: adult. Fig. 23. *Liriomyza lathyri*: completed and vacated mine. Fig. 24. *Liriomyza* sp. prob. *robiniae*: vacated mine. Fig. 25. *Nemorimyza posticata*: vacated mine showing arced feeding lines.
(Yu et al. 2012, Noyes 2018). **Discussion:** The completed mine is a full depth blotch. The frass is scattered irregular-sized black particles. The larva exits its mine at the leaf edge prior to pupating. Though no adults were obtained, this identification is based upon host, early seasonal occurrence of mine, and mining characters described and illustrated as an unknown Agromyzidae by Weaver and Dorsey (1967). Valley (1982) subsequently referred to the mining characters in Weaver and Dorsey (1967) as *L. robitiae* after a conversation of Valley with Weaver. *Liriomyza robiteiae* is the only known Agromyzidae leaf miner of *R. pseudoacacia.*

**Nemorimyza posticata** (Meigen) (Fig. 25). **Specimen data:** Quarry-Fen 46°53.839N 87°53.607W, 11 larvae 18 Jul 2003, 1♂ 07 Aug 2003, Lot 1424; Loop Rd. 46°50.627N 87°51.300W, 8 larvae 20 Sep 2007, 3 adults after winter, Lot 1843. **Distribution:** COSTA RICA, EUROPE, JAPAN, CANADA: AB; U.S.A.: Probably all States (Sehgal 1971, Spencer and Steyskal 1986). **Parasitoids:** *Braconidae:* *Chorebus* (Stiphrocerca) anasellus (Stelfox). *Dacnusa* (*Pachysema*) discolor, *Dacnusa* (*Dacnusa*) maculipes, *Dacnusa* (*Dacnusa*) plantaginis Griffiths, *Dacnusa* (*Pachysema*) sibirica Telenga, *Dacnusa* (*Pachysema*) zlobini Tobias, *Dapsilarthra* (*Dapsilarthra*) rufiventris, *Opia* (*Gastrosema*) osecindis (Ashmead), *Phaedrotoma* depucerator Stein., *Phaedrotoma* diverse (Szépligeti), *Phaedrotoma* exigua (Wesmael) (Yu et al. 2012); *Eulophidae:* *Chrysocharis* pubicorris, *Chrysocharis* viridis, *Diglyphus isaea*, *Hemiptarsenus ornatus* (Nees), *N. gatuloticornis* L.; *Pteromalidae:* *Halticoptera aenea* (Walker); *Tetracampidae:* *Epiclerus panyas* (Walker) (Noyes 2018). **Remarks:** One or two mines occur per leaf, but there is only one larva per mine. Each mine is confined to one side of the main vein. When fully formed the mine is an upper surface elongate blotch with a rounded herringbone feeding pattern evident. Only a few scattered black frass particles are visible. The larva exits the upper leaf surface prior to pupating. From these emergence records there appears to be two generations per year at HMC.

**Phytomyza loewii** Hendel (Fig. 26, 27). **Specimen data:** Ives Lake Rd. 46°50.475N 87°50.581W, 6 larvae 26 Sep 2002, 1♂ after wintering, Lot 1357. **Distribution:** CANADA, U.S.A.: CA, DC, GA, ID, IN, LA, WA (Spencer and Steyskal 1986). **Parasitoids:** *Cleomis* spp., *C. ligusticifolica* Nutt. and *C. verticillaris* DC. (Spencer and Steyskal 1986). **Remarks:** One or two mines occur per leaf, but there is only one larva per mine. The completed mine is linear on the upper surface with frass in a dark trail along one side of the mine. The larva exits the lower leaf surface prior to pupating. *Dacnusa discolor* is currently known only from the Palearctic Region. It has been reported from Russia east to Azerbaijan (Yu et al. 2012). If verified the specimens in this research would be the first record of this species for the Near World and the first braconid reported from *P. loewii.*

**Phytomyza sp. prob. plantaginis Robin-Besa (Fig. 28). **Specimen data:** Case residence 46°50.892N 87°49.783W, 1 puparium 17 Jul 2003, 0 adult, Lot 1419. **Distribution:** CANADA: AB, ON, QC (Spencer 1969, Sehgal 1971); U.S.A.: “Widespread through most of United States” (Spencer and Steyskal 1986). **Parasitoids:** *Braconidae:* *Chorebus* (Stephrocerca) anasellus (Stelfox). *Dacnusa* (*Pachysema*) discolor, *Dacnusa* (*Dacnusa*) maculipes, *Dacnusa* (*Dacnusa*) plantaginis Griffiths, *Dacnusa* (*Pachysema*) sibirica Telenga, *Dacnusa* (*Pachysema*) zlobini Tobias, *Dapsilarthra* (*Dapsilarthra*) rufiventris, *Opia* (*Gastrosema*) osecindis (Ashmead), *Phaedrotoma* depucerator Stein., *Phaedrotoma* diverse (Szépligeti), *Phaedrotoma* exigua (Wesmael) (Yu et al. 2012); *Eulophidae:* *Chrysocharis* pubicorris, *Chrysocharis* viridis, *Diglyphus isaea*, *Hemiptarsenus ornatus* (Nees), *N. gatuloticornis* L.; *Pteromalidae:* *Halticoptera aenea* (Walker); *Tetracampidae:* *Epiclerus panyas* (Walker) (Noyes 2018). **Remarks:** One or two mines occur per leaf, but there is only one larva per mine. Each mine is confined to one side of the main vein. When fully formed the mine is an upper surface elongate blotch with a rounded herringbone feeding pattern evident. Only a few scattered black frass particles are visible. The larva exits the upper leaf surface prior to pupating. From these emergence records there appears to be two generations per year at HMC.

**Phytomyza loewii** Hendel (Fig. 26, 27). **Specimen data:** Ives Lake Rd. 46°50.475N 87°50.581W, 6 larvae 26 Sep 2002, 1♂ after wintering, Lot 1357. **Distribution:** CANADA, U.S.A.: CA, DC, GA, ID, IN, LA, WA (Spencer and Steyskal 1986). **Parasitoids:** *Cleomis* spp., *C. ligusticifolica* Nutt. and *C. verticillaris* DC. (Spencer and Steyskal 1986). **Remarks:** One or two mines occur per leaf, but there is only one larva per mine. The completed mine is linear on the upper surface with frass in a dark trail along one side of the mine. The larva exits the lower leaf surface prior to pupating. *Dacnusa discolor* is currently known only from the Palearctic Region. It has been reported from Russia east to Azerbaijan (Yu et al. 2012). If verified the specimens in this research would be the first record of this species for the Near World and the first braconid reported from *P. loewii.*

**Phytomyza sp. prob. plantaginis Robin-Besa (Fig. 28). **Specimen data:** Case residence 46°50.892N 87°49.783W, 1 puparium 17 Jul 2003, 0 adult, Lot 1419. **Distribution:** CANADA: AB, ON, QC (Spencer 1969, Sehgal 1971); U.S.A.: “Widespread through most of United States” (Spencer and Steyskal 1986). **Parasitoids:** *Braconidae:* *Chorebus* (Stephrocerca) anasellus (Stelfox). *Dacnusa* (*Pachysema*) discolor, *Dacnusa* (*Dacnusa*) maculipes, *Dacnusa* (*Dacnusa*) plantaginis Griffiths, *Dacnusa* (*Pachysema*) sibirica Telenga, *Dacnusa* (*Pachysema*) zlobini Tobias, *Dapsilarthra* (*Dapsilarthra*) rufiventris, *Opia* (*Gastrosema*) osecindis (Ashmead), *Phaedrotoma* depucerator Stein., *Phaedrotoma* diverse (Szépligeti), *Phaedrotoma* exigua (Wesmael) (Yu et al. 2012); *Eulophidae:* *Chrysocharis* pubicorris, *Chrysocharis* viridis, *Diglyphus isaea*, *Hemiptarsenus ornatus* (Nees), *N. gatuloticornis* L.; *Pteromalidae:* *Halticoptera aenea* (Walker); *Tetracampidae:* *Epiclerus panyas* (Walker) (Noyes 2018). **Remarks:** One or two mines occur per leaf, but there is only one larva per mine. Each mine is confined to one side of the main vein. When fully formed the mine is an upper surface elongate blotch with a rounded herringbone feeding pattern evident. Only a few scattered black frass particles are visible. The larva exits the upper leaf surface prior to pupating. From these emergence records there appears to be two generations per year at HMC.

**Phytomyza loewii** Hendel (Fig. 26, 27). **Specimen data:** Ives Lake Rd. 46°50.475N 87°50.581W, 6 larvae 26 Sep 2002, 1♂ after wintering, Lot 1357. **Distribution:** CANADA, U.S.A.: CA, DC, GA, ID, IN, LA, WA (Spencer and Steyskal 1986). **Parasitoids:** *Cleomis* spp., *C. ligusticifolica* Nutt. and *C. verticillaris* DC. (Spencer and Steyskal 1986). **Remarks:** One or two mines occur per leaf, but there is only one larva per mine. The completed mine is linear on the upper surface with frass in a dark trail along one side of the mine. The larva exits the lower leaf surface prior to pupating. *Dacnusa discolor* is currently known only from the Palearctic Region. It has been reported from Russia east to Azerbaijan (Yu et al. 2012). If verified the specimens in this research would be the first record of this species for the Near World and the first braconid reported from *P. loewii.*

**Anthomyiidae**

*Chirocephlosia spinosissima* (Malloch) (Fig. 29, 30). **Specimen data:** Near Jensen homestead, 46°53.656N 87°53.131W, 7 larvae, 16-17 Jul 2003, 1♂ after wintering, Lot 1417. **Distribution:** CANADA: NB, ON, QC;
Figures 26–33. **Fig. 26.** *Phytomyza loewii*: adult. **Fig. 27.** *Phytomyza loewii*: vacated mine. **Fig. 28.** *Phytomyza* sp. prob. *plantaginis*: vacated mine. **Fig. 29.** *Chirosia spinosissima*: adult. **Fig. 30.** *Chirosia spinosissima*: lower mine surface with external frass trail between adjacent mined pinnae. **Fig. 31.** *Chirosia* species: larva feeding toward base of pinna. **Fig. 32.** *Pegomya rumicifoliae*: adult. **Fig. 33.** *Pegomya rumicifoliae*: single mine with larva.
U.S.A.: MI, NC, WI (Griffiths 2004). Larval hosts: None reported in Griffiths (2004). *Pteridium aquilinum* (L.) Kuhn (identified in this survey). Parasitoids: None reported (Yu et al. 2012, Noyes 2018). Pteromalidae: *Lamprotatus* sp.; Braconidae: *Utetes (Utetes) parvifossa* (Fischer) (identified in this survey). Remarks: The full depth mine includes part or an entire frond pinna. The frass is dark green forming a central trail. The larva exits its mine via the lower surface. The recovered larva was nearly mature, and it was apparent the mine size was too small to account for its entire development. The larva likely exited its initial mine and entered another pinna to continue feeding. This is supported by a frass trail on the lower surface between two partially mined pinnae. With only one cut per mine, it is also apparent the larva enters and exits its mine through this single cut. This is the first report of a larval host for this species though adults have been reported in Michigan (Griffiths 2004). Griffiths suggested larvae would be found on ferns. In his concept of the genus, *Chriosia* species are the only known Anthomyiidae with larvae mining *Pteridium* species. *Lamprotatus* sp. is the first chalcidoid reported from *C. spinosissima*.

*Chriosia* sp. (Fig. 31). Specimen data: Jensen homestead 46°53.656N 87°53.131W, 1 larva 14 Sep 2006, 0 adult, Lot 1750. Larval host: *Pteridium aquilinum* (identified in this survey). Parasitoids: None reported (Yu et al. 2012, Noyes 2018). Braconidae: *Utetes (Utetes) parvifossa* (identified in this survey). Remarks: This single specimen was found at the same site as *C. spinosissima*, which has a similar mine and frass pattern, though occurring later in the season. The larva exited the lower mine surface prior to pupating as does *C. spinosissima*. An issuing adult would likely have emerged after winter as is the case with *C. spinosissima* and would agree with Griffiths (2004) records of *C. spinosissima*. All 16 *Chriosia* species listed by Griffiths (2004) in northeastern North America have one generation per year with adults issuing between April and June. Fifteen of these species have no recorded larval host. The single larval specimen documented here may be *C. spinosissima*, though its two month later appearance, compared with the earlier confirmed rearing, makes one of the other 15 species, without known hosts, possible candidates.

*Pegomya rumicifoliae* Hewckett (Fig. 32, 33). Specimen data: Loop Rd. 46°50.627N 87°51.300W, 11 larvae 06 Jun 2006, 7♂, 2♀ 27-28 Jun 2006, Lot 1699. Distribution: CANADA: AB, ON, QC, U.S.A.: MA, MI, NJ, NY, RI, WI (Griffiths 1982), ME (Eiseman 2018). Larval hosts: *Rumex crispus* L., *R. obtusifolius* L., *R. mexicanus Meisn.* (Griffiths 1982), *R. britannica* L. (Eiseman 2018). *Rumex obtusifolius* (identified in this survey). Parasitoids: None reported (Yu et al. 2012, Noyes 2018). Remarks: There are frequently several mines per leaf, each forming a widening track. Mines eventually coalesce forming a single mine over much of the upper leaf surface. Each initial mine has an opening at its base. Frass, which is composed of irregular black particles, is deposited at the mine center as an intermittent winding trail. Frost (1924) (as *P. calyptrata* (Zetterstedt)) and later Eiseman (2018) reported multiple generations per year; only one was observed and reared in this survey.

**HYMENOPTERA**

**Tenthredinidae**

*Fenella nigrita* Westwood (Fig. 34, 35). Specimen data: Loop Road 46°50.627N 87°51.300W, 21 larvae 20 Sep 2007, 1♂, 08 Oct 2007, Lot 1846. Distribution: EUROPE; CANADA: ON; U.S.A.: CT, ME, MI (Smith 1971). Larval hosts: *Potentilla* sp. (Smith 1971). *Potentilla norvegica* (identified in this survey). Parasitoids: None reported (Noyes 2018); Braconidae: *Colastes (Shawiana) catenator* (Haliday), *Colastes (Xenarcha) lustrator* (Haliday); Ichneumonidae: *Hemiteles pygmaeus* Brischke, *Perillus cingulatus* Brischke, *Perillus fenellae* Brischke (Yu et al. 2012). Remarks: Only one mine was found per leaflet and one larva per mine. The larva feeds ventral side up. The mine is full depth, beginning as a widening track then later broadening into a blotch. Frass is very distinct black particles initially placed in a wide central trail then later scattered in the blotch portion.

pictus (Nees), Cirrospilus viattus Walker, Closterocerus trifasciatus Westwood, Hemiptarsenus ornatus, Minotetrastichus frontal (Nees), Pnigalio cruciatilus (Ratzelburg), Pnigalio maculipes (Crawford), Pnigalio minio (Walker), Sympiesis sp., Sympiesis gordus (Walker), Sympiesis sericeornis (Nees); Torymidae: Monodontomerus indiscreetus Gahan; Trichogrammatidae: Trichogramma minutum (Nees); gordius minio (Crawford), Pnigalio maculipes (Nees), tarsenus ornatus Gnathochorisis dentifer foliicola Cushman, Mason, laplantei Exochus pictus xanthopsis Ashmead, Townes and Townes, pon metallicum Hoebeke and Wheeler, Jr. (2005) reported larva of the third species, to pupating. The larva exits the upper leaf surface prior to pupating. Black frass particles are concentrated is blotch-shaped initiated away from the leaf edge. Frass is composed of scattered black oblong particles. The dearth of frass inward. Frass is scattered throughout the leaf and visible from the lower surface. The larva usually exits its mine via the lower leaf surface though occasionally from the upper surface. The larva pupates in its mine by forming a circular cocoon approximately 8 mm in diameter. Both mine and adult agree with the descriptions provided by Digweed et al. (2009).

Metallus sp. prob. capitalis (Norton) (Fig. 38). Specimen data: Loop Road 46°50.627N 87°51.300W, 3 larvae 04 Oct 2009, 0 adults, Lot 1958. Distribution: Probably transcontinental across northern United States and southern Canada. CANADA: BC, NB, NL, NS, ON, QC; U.S.A.: IA, ID, IL, ME, MA, MI, NH, NY, OR, VT (Eiseman and Smith 2017). Larval hosts: Rubus sp. (Smith 1971). Remarks: There is one mine per leaflet with one to two miners per mine. The mine is a full depth blotch, reddened at its leaf edge origin and lightening inward. Frass is composed of scattered black oblong particles. The deth of frass and absence of cast larval exuvia imply most was expelled through the slit located in the leaf edge. The larva pupates in its mine by forming a circular cocoon approximately 8 mm in diameter. Both mine and adult agree with the descriptions provided by Digweed et al. (2009).

Metallus sp. prob. rohweri MacGillivray (Fig. 39). Specimen data: Howe Lake 46°53.823N 87°57.668W, 11 larvae 25 Sep 2002, 0 adults, Lot 1356; Flat Rock 46°54.520N 87°55.355W, 16 larvae 19 Sep 2003, 0 adults, Lot 1444; Rush Lake Boat House 46°53.208N 87°53.681W, 43+ larvae 14 Sep 2005, 0 adults, Lot 1655. Distribution: CANADA: NB, NS, ON; U.S.A.: CT, DE, FL, IL, ME, MD, MA, MI, MO, NJ, NY, NC, OH, PA, RI, VA (Smith 1971). Larval hosts: Rubus sp. (Smith 1971); R. canadensis L. (Lot 1655), R. flagellaris Willd. (Lot 1356), and R. parviflorus Nutt. (Lot 1444) (identified in this survey). Parasitoids: Braconidae: Colastes (Shawiana) metalli (Muesebeck), Proterops proteroptoides (Viereck); Ichneumonidae: Campoletis argentifrons (Cresson), Endasys praerotundiceps Luhman, Isorropus coelebs (Walsh), Lathristes (Lathrosthes) constrictus (Provancher), Lathrosthes (Lathrosthes) truncatus (Provancher), Scambus hispas (Yu et al. 2012). Euophila: Pnigalio maculipes, P. minio (Noyes 2018). Remarks: Eggs are imbedded in the leaf and visible from the lower surface. Mines are full depth becoming near a vein as a widening track then later expanding into a swollen blotch that may coalesce with other mines. Five to seven miners occur per leaflet with one to two miners per mine. Frass is both clumped and scattered. The larva usually exits its mine via the lower leaf surface though occasionally from the upper surface. These observations agree with those provided by Daniel (1928) except for the larva exiting its mine primarily from the lower surface. Daniel (1928) described two generations per year in western New York State, but only one generation was observed during this survey. The single generation per year agrees with observations of Eiseman and Smith (2017).

Nefusa sp. prob. ambigua (Norton) (Fig. 40). Specimen data: Loop Road 46°50.627N 87°51.300W, 3 larvae 20 Sep 2007, 0 adults, Lot 1847. Distribution: CANADA: NS, QC; U.S.A.: AR, IA, IL, KY, MA, MD, MA, MI, MO, NY, OH, PA, TN, TX, WV, VA, WI (Smith and Eiseman 2015). Larval hosts: Viola canadensis L., V. palmata L., V. pubescens Ait., V. sororia Willd. (Smith and Eiseman 2015). Viola sp. (identified in this survey). Parasitoids: None reported (Yu et al. 2012, Noyes 2018). Remarks: Only one mine per leaf and one larva per mine were observed. The mine is full depth. It likely begins at the leaf edge though the early mine area was dried and wrinkled when recovered. The completed mine encompassed much of the leaf. Frass initially appears as a brown smear and later becomes distinct black particles arranged in an intermittent line. The larva exits the
Figures 34–41. Fig. 34. *Fenella nigrita*: adult. Fig. 35. *Fenella nigrita*: near-mature larva feeding ventral side up. Fig. 36. *Heterarthrus nemoratus*: adult. Fig. 37. *Heterarthrus nemoratus*: nearly completed mine with feeding larva. Fig. 38. *Metallus* sp. prob. *capitalis*: completed mine. Fig. 39. *Metallus* sp. prob. *rohweri*: 3 mines on *Rubus canadensis*. Fig. 40. *Nefusa* sp. prob. *ambigua*: nearly completed mine with larva. Fig. 41. *Profenusa* sp. prob. *alumna*: upper leaf surface with larva feeding, prolegs visible.
lower leaf surface prior to pupating. These observations agree with the brief notes of Shaw (1940) and the more extensive observations of Smith and Eiseman (2015). This is the only species of Tenthredinidae in North America with larvae known to feed in Viola.

_Profensusa_ sp. _prob. alurna_ (MacGillivray) (Fig. 41). **Specimen data:** Rush Lake Boat House 46°53.209N 87°54.631W, 4 larvae 14 Sep 2006, 0 adults, Lot 1752. **Distribution:** U.S.A.: IL, ME, MD, NY, PA, VA (Smith 1971). **Larval hosts:** _Quercus alba_ L., _Q. ilex_ (Watson.), _Q. macrolepida_ Michx., _Q. rubra_ _Q. velutina_ Lam. (Smith 1971). _Quercus rubra_ (identified in this survey). **Parasitoids:** _Ichneumonidae:_ *Lathrolestes* (Lathrolestes) _luteolator_ (Gravenhorst). _Lathrolestes* (Lathrolestes) _platygynus_ (Davis) (Yu et al. 2012). **Remarks:** One mine per leaf and one larva per mine were observed. The upper surface mine begins as a widening track becoming a blotch. The larva feeds ventral side upward and exits the upper leaf surface prior to pupating. Black frass particles are scattered within its mine though away from the mine perimeter. _Profensusa _is the only genus of Tenthredinidae known to mine _Quercus_ leaves. Of the five species in North America, three are miners of _Quercus_ leaves.

_Profensusa_ _prob. thomsoni_ (Konow) (Fig. 42). **Specimen data:** Loop Road 46°50.627N 87°51.300W, 2 larval skins 27 Sep 2002, 0 adult, Lot 1365; Quarry-Fen 46°53.839N 87°53.607W, 1 larva 17 Sep 2003, 0 adults, Lot 1441; same location, 2 larvae 16 Aug 2004, 0 adults, Lot 1515; Gillet Landing 46°51.870N 87°46.435W, 7 larvae 16 Aug 2004, 0 adults, Lot 1510; Picnic Point 46°55.056N 87°50.506W, 2 vacated mines 12 Sep 2005, Lot 1645; Rush Lake Boat House 46°53.209N 87°54.631W, 2 larvae 14 Sep 2006, 0 adults, Lot 1751. **Distribution:** EUROPE: CANADA: ON, QC, U.S.A.: CT, ME, VT (Smith 1971). **Larval hosts:** _Betula alleghaniensis, B. papyrifera_, and _B. populifera_ (Smith 1971). _Betula papyrifera_ (identified in this survey). **Parasitoids:** _Eulophidae:_ *Chrysocharis* _erumpens_ Graham. _Chrysocharis nesperus, Chrysocharis nitetis, Chrysonotomia sp., Minotetrauchis sp., Pnigalio sp., Sympiesis sp., Zagrammosoma sp._ **Trichogammatidae:** _Trichogamma_ _ausoma_ Sugonjaev and Sorokina (Noyes 2018). _Ichneumonidae:_ *Lathrolestes* (Lathrolestes) _luteolator, Lathrolestes* (Lathrolestes) _soperi_ Reschikov, _Lathrolestes* (Lathrolestes) _thomsoni_ Reschikov (Yu et al. 2012). **Remarks:** One or two mines may occupy a leaf but only a single larva per mine. The upper surface mine begins as a widening track and later becomes a blotch. Frass is initially clumped forming a broken trail then later scattered. The larva exits the upper leaf surface at or near the mine margin prior to pupating. Mines of reared lots easily key to _P. thomsoni_ in Digweed et al. (2009) who include all known species in Canada.

**LEPIDOPTERA**

*Argyresthiidae*

_Argyresthia thuiella_ Packard (Fig 43, 44). **Specimen data:** Gillet Landing 46°51.870N 87°46.435W, 7 larvae 11 Jun 2001, 3♂, 27-28 Jun 2001, Lot 1127. **Distribution:** CANADA: Eastern Canada; (Freeman 1972), U.S.A.: ME, PA (Busck 1907); MI (Nielsen 1998). **Larval hosts:** _Thuja occidentalis_ L. (Freeman 1972). _Thuja occidentalis_ (identified in this survey). **Parasitoids:** *Braconidae:* _Apanteles_ (Apanteles) _epinotia_ (Viereck), _Apanteles_ (Apanteles) _paralechiae_ Muesebeck, _Apanteles_ (Apanteles) _thujae_ Muesebeck, _Bracon_ (Bracon) _speerschneideri_ Schmiedeknecht, _Charmion_ (Charmion extensor) (Linnaeus), _Chelonus_ (Microchelonus) _erucariae_ McComb, _Hypomicrogaster_ _zoniae_ (Say), _Pholetesor bedelliae_ (Viereck), _Pholetesor thuiellae_ Whiffin. _Ichneumonidae:_ _Pori- zon cupressi_ (Ashmead) (Yu et al. 2012). _Chalcididae:_ _Conura albibrons, C. side_ (Walker); _Encytiidae:_ _Copidosoma bucculatrix_ (Howard); _Eulophidae:_ _Baryscapus coleulescens_ (Ashmead), _Cirsospilus _vittatus, _Clostrecerus triflascus, Dicladocerus_ _spherocerus, Dicladocerus vulgaris_ Yoshimoto, _Euderus_ _cushmanii_ (Crawford), _Necremnus_ _sp., Neo- chrysocharis formosa_ (Westwood), _Pediobius alipes, Pnigalio sp._, _Pnigalio maculipes, Pnigalio minio, Symphyesis _stigmatipennis_ Girault; _Eupelmidae:_ _Eupelmus vesicularis_ (Retzus) (Noyes 2018). _Eulophidae:_ _Pnigalio maculipes, Symphyesis _sericeicornis_ (identified in this survey). **Remarks:** The solitary larva consumes all green tissue throughout the mined area though that area remains opaque. Pupation occurs within the mine. This is a new host record for _Symphyesis sericeicornis_.

*Bucculaticridae*

_Bucculatrix canadensisella_ Chambers (Fig. 45, 46). **Specimen data:** Salmon Trout Bay 46°51.870N 87°46.435W, 8 larvae 25-26 Sep 2000, 1♂ after wintering. Lot 1082, same site, 14+ larvae 16 Sep 2006, 3♂, 6♀ after wintering. Lot 1758; same site, 1 larva 19 Sep 2007, 1♂ after wintering. Lot 1849. **Distribution:** CANADA: AB, BC, MB, NB, NS, ON, PE, QC, SK; U.S.A.: CO, KY, MI, MN, NJ, NY, NC, PA, TN, WI (Friend 1927, Braun 1963). **Larval hosts:** _Betula lenta_ L., _B. lutea_ Michx., _B. nigra, B. occidentalis, B. papyrifera, B. populifolia_ (Braun 1963). _Betula papyrifera_ (identified in this survey).
Figures 42–49. Fig. 42. Profenusa sp. prob. thomsoni: completed mine. Fig. 43. Argyresthia thuiella: adult. Fig. 44. Argyresthia thuiella: completed mine. Fig. 45. Bucculatrix canadensisella: adult. Fig. 46. Bucculatrix canadensisella: late instar larva skeletonizing lower leaf surface. Fig. 47. Bucculatrix packardella: adult. Fig. 48. Coleophora pruniella: adult. Fig. 49. Coleophora pruniella: lower leaf surface with circular entrance in mine.
Parasitoids: Braconidae: Cantharotoncus canadensis Mason, Pholetesor bedelliaria, Pholetesor thuellia, Stiropius bucculaticis; Ichneumonidae: Campoplex brachyurus Viereck. Gelis obscurus, Gelis urbanus (Yu et al. 2012). Chalcididae: Haltichella xanticles (Walker); Eulophidae: Chrysoscharis nephereus, Cirrospilus cinctithorax, Cirrospilus ocellatus Girault, Closterocerus cinctipennis, Derostenus sp., Pediobius bucculaticis (Gahan) (Noyes 2018). Braconidae: Stiropius bucculaticis; Eulophidae: Pediobius albipes (identified in this survey). Remarks: Though the early instar larva is a miner, the later instar is a lower leaf surface skeletonizer occasionally feeding on the upper surface. The larva is pale yellow with white pinacula and brown setae. The cocoon is dark brown with six ribs. This is a new host record for *Pediobius albipes* (identified in this survey).

Though Mesochorus americanus Cresson, M. parvus Dasch, and M. vittator (Zetterstedt) are listed as parasitoids in Yu et al. (2012), they are known to be hyperparasitoid (RRK).


**Parasitoids:** Eulophidae: *Pnagio maculipes* (Noyes 2018). Remarks: The nearly fully developed larva skeletonizes the lower leaf surface. The single cocoon recovered was white with seven to eight ribs. Braun (1963) states that 10 – 12 ribs are usual.

**Coleophoridae**


**Parasitoids:** Chalcididae: *Conura* sp., *Conura albifrons*, *Conura side; Encyrtidae: Copidosoma truncatellum* (Dalman). Eulophidae: *Chrysoscharis* sp., *Chrysoscharis ainsliei; C. iaricinellae* (Ratzeburg), *C. pentheus*, *Cirrospilus* sp., *Cirrospilus cinctithorax*, *C. flavicinctus*, *Closterocerus* sp., *Closterocerus trifasciatus*, *Derostenus* sp., *Elachertus* sp., *Elachertus fenestratus* Nees, Elasmus setosiscutellatus Crawford, Euderus cushmanii, Eulophus sp., *Eulophus magnisulcatus* Girault, *Horismenus* sp., *Horismenus fraternus* (Fitch), Minotetrastichus frontalis, Pnagio sp., P. maculipes, P. minio, P. pallipes (Provancher), *Sympeesia* sp., *S. sericeicornis*, *S. stigma*ta Girault, *Tetrastichus* sp., *Zagrommosoma* sp.; Eupelmidae: *Eupelmus vesicularis*; Eurytomidae: *Eurytoma appendigaster* (Swederus), *E. verticillata* (Fabricius); Pteromalidae: *Catolaccus aeneoviridis* (Girault), *Catolaccus kansensis* (Girault), *Hypopteromalus inimicus* Muesebeck, Hypopteromalus perssirius* (Girault), *Pteromalus* sp., *P. phycidis* (Ashmead), *P. thyridopteris* (Howard), Sceptrothelys deione (Walker), *Trichomalopsis* sp.; Torymidae: *Torymus* sp.; Tricogrammatidae: *Trichogramma minutum* (Noyes 2018). Braconidae: *Braco (Braco) pygmaeus* Provancher, *Habrobracon geleiidae* (Ashmead); Ichneumonidae: *Campoplex mellipes* (Provancher), *Diphyus comes* (Cresson), *Gelis tenellus* (Say), *Ichneunon annulariorus* Fabricius, *Ilopectis conquisitor* (Say), Ilopectis quadriringulata (Provancher), *Seambus calobatus* (Gravenhorst), *Scambus decorus* Walley, *Scambus hispae*, *Spilichneumon inconstans* (Cresson) (Yu et al. 2012). Remarks: One case was recovered affixed to the upper surface of a *Crataegus* leaf. Several scattered blotch mines were present on adjacent leaves. Mines contained no frass, but each had a circular hole in the center of the mine on the lower surface characteristic of coleophorid larval feeding. The initial portion of the cases is light tan while the later portion is darker.

**Eriocraniidae**

*Eriocrania* sp. prob, *semipurpurella* Stephens (Fig. 50). Specimen data: Picnic Point 46°53.056N 87°50.509W, 3 live larvae 05 Jun 2006, no adults, Lot 1697; Upper Falls Road 46°50.627N 87°51.300W, 3 mines, 2 with dead larvae, third vacant 08 Jun 2006, 0 adults, Lot 1703. Distribution: Great Britain, northern and central Europe, Japan; CANADA: NS, ON, QC, U.S.A.; NY (Davis 1978). Larval hosts: *Betula pendula* Roth, *B. pubescens* Ehrh., *B. platyphylla* Sukaczew var. *japonica* (Miq.) Hara (hosts not from North America) (Davis 1978). *Betula papyrifera* (Lot 1697) and *B. alleghaniensis* (Lot 1703) (identified in this survey). Parasitoids: Braconidae: *Colastes* (Shawiana) catenator. Ichneumonidae: *Cryptocerus basalis* Ruthe, *Lathrolestes* (Lathrolestes) clypeatus (Zetterstedt) (Yu et al. 2012); Eulophidae: *Aprostocetus pallipes* (Dalman), *Chrysoscharis nephereus*, *Cirrospilus lyncus* Walker, *C. pictus*, *Closterocerus trifasciatus*, *Minotetrastichus frontalis*, *Pnagio eriocraniae* Li and Yang, *P. longulus* (Zetterstedt), *P. soemius* (Yu et al. 2012). Remarks: There are one to two mines per leaf with a single...
larvae larva in each. The full depth mine is initiated at the leaf edge, developing as a widening track. Frass at the mine origin is densely packed, but as the mine expands frass clearly appears as long black strings. Prior to pupating the larva exits its mine through either the upper or lower leaf surface. Preserved larvae were identified to family (Davis 1987). Davis lists various Betula species as hosts in the Palearctic though none in North America. The mine characteristics described and illustrated here agree with the image of this species on the website http://www.leafmines.co.uk/ (2009).

Gelechiidae

Scrobipalpula manierreorum Priest (Fig. 51, 52). Specimen data: Loop Road 46°50.627N 87°51.300W, 6 larvae 17 Aug 2007, 0 adults, Lot 1523; same site, 7 larvae 13 Sep 2005, 4 adults after wintering, Lot 1654; same site, 3 mines 14 Sep 2006, 0 adults Lot 1763; same site, 4 larvae 28 Aug 2007, 3 adults after wintering, Lot 1835; same site, 2 larvae 27 Aug 2008, 2 adults after wintering. Lot 1872; Upper Falls Road 46°50.475N 87°50.581W, 2 mines 17 Sep 2006, 1 adult, Lot 1768; Ivies Lake Rd. at Club Rd. 46°50.926N 87°48.012W, 9 larvae 28 Aug 2007, 1 adult after wintering, Lot 1834; Quarry-Fen 46°53.839N 87°53.607W, 20 mines 28 Aug 2010, 2 adults after wintering, Lot 1889. Distribution: CANADA: AB, BC, MB, ON, QC; U.S.A.: MI (Adamski et al. 2014). Larval hosts: Eurybia (Aster) macrophylla (Adamski et al. 2014). Parasitoids: Braconidae: Schoenlandella minuta (Cresson); Ichneumonidae: Campoplex sp. (Adamski et al. 2014). Remarks: One to 14 mines occur per leaf though miners are solitary. The mine usually begins at the main vein extending toward the leaf margin as a full depth narrow branching tract. Frass is fastened with silk externally on the lower surface at the mine base forming a curved tube. It is used as a larval retreat if disturbed or not feeding. Frass is also placed in the mine as double rows, which serve to guide the larva backward to its frass tube based on RJP observations. When feeding is complete the larva exits its mine through its frass tube to winter on the ground. There was only a single generation observed in the study. Most of the above data was originally published in Adamski et al. (2014).

Scrobipalpula sacculicola (Braun) (Fig. 53, 54). Specimen data: Loop Road 46°50.627N 87°51.300W, 1 larva 20 Sep 2007, 1 ♀ after wintering. Lot 1844. Distribution: CANADA: ON, QC; U.S.A.: CO, LA, MI, OH, OK (V. Nazari, personal communication). Larval hosts: Solidago sp. (Braun 1925). Solidago sp. (identified in this survey). Parasitoids: None reported (Yu et al. 2012, Noyes 2018). Remarks: The single mine recovered contained one larva. The full depth mine begins basally between the main vein and a secondary vein. It develops as a straight gradually expanding track along one side of the main vein toward the leaf apex. Frass is minute, black, and sparse. Most frass is expelled through a hole in the mine floor at its origin.

Gracillariidae


Eulophidae: Pedioibius alcaeus (Walker); Braconidae: Colastes sp. 1, Pholetesor (?) (identified in this survey). Remarks: There are one to four mines per leaf but only one miner in each. Initially the mine is serpentine later widening into a blotch. The initial portion is usually clearly visible on the upper leaf surface, but upon expanding the mine may be full depth or meander between leaf surfaces. Frass in the serpentine part is a light colored central trail but with clearly scattered black particles in the blotch portion. The feeding larva is tan colored, but upon exiting its mine, via the upper leaf surface, its color turns pink. The cocoon is pale pink and approximately 5.5 x 1.4 mm. The specimens of Colastes and Pholetesor are the first species of Braconidae reported as parasitic on A. astericola. This is a new host record for Pedioibius alcaeus.

Cameraria aceriella (Clemens) (Fig. 57, 58). Specimen data: Howe Lake 46°53.823N 87°57.668W, 5 larvae 28 Sep 2000, 1♂ after wintering, Lot 1098; Quarry-Fen 46°53.839N 87°53.607W, 5 larvae 17 Sep 2003, 1♂, 2♀ after wintering. Lot 1438; Flat Rock 46°54.520N 87°55.355W, 5 larvae 12 Sep 2005, 0 adults, Lot 1647. Distribution: CANADA: QC; U.S.A.: CT, IL, KY, ME, MD, MI, NY, PA, VT, WI (De Prins and De Prins 2005). Larval hosts: Acer rubrum L., A. saccharinum L., A. saccharum (De Prins and De Prins 2005). Acer saccharum (identified in this survey). Parasitoids:
Figures 50–57. Fig. 50. *Eriocrania* sp. *prob. semipurpurella*: larva feeding on *Betula papyrifera*. Fig. 51. *Scrobipalpula manierreorum*: adult male, holotype. Fig. 52. *Scrobipalpula manierreorum*: completed mine reaching leaf edge with curved basal frass tube visible on lower surface. Several additional mines are present, mostly initiated along main vein. Fig. 53. *Scrobipalpula sacculicola*: adult. Fig. 54. *Scrobipalpula sacculicola*: mine with feeding larva. Fig. 55. *Acrocercops astericola*: adult. Fig. 56. *Acrocercops astericola*: completed mine. Fig. 57. *Cameraria acriella*: adult.
**Eulophidae**: *Chrysocharis occidentalis* (Girault), *Pediobius alceaeus*, *Pnigalio pallipes* (Noyes 2018); **Braconidae**: *Centistidea* (Centistidea) *lithocellitidis* (Ashmead), *Pholetesor ornigis* (Weed); **Ichneumonidae**: *Alophosternum folicola* (Yu et al. 2012).

**Remarks**: One to three mines occur per leaf but each with only a single miner. The mine is a wide track at times recurving to form an apparent blotch. Black frass forms a narrow trail along the mine perimeter. Pupation occurs in the mine. The larva forms an approximately 6.0 mm white cocoon at its mine terminus.

**Cameraria corylisella** (Chambers) (Fig. 62, 63). **Specimen data**: Loop Road 46°50.627N 87°51.300W, 5 completed mines 13 Sep 2005, 1♂ after wintering, Lot 1649.

**Distribution**: **CANADA**: QC; **U.S.A.**: CT, IL, KY, ME, NY, VT, WI (De Prins and De Prins 2005). **Larval hosts**: *Coriulys americana* Walt., *Ostrya virginiana* (Miller) K. Koch, *Carpinus americana* Walt., *C. caroliniana* Walt. (De Prins and De Prins 2005), *Corylus cornuta* Marsh. (identified in this survey). **Parasitoids**: **Eulophidae**: *Pediobius ocellatus* Peck; **Braconidae**: *Pholetesor ornigis*; **Ichneumonidae**: *Alophosternum folicola* (Yu et al. 2012) (identified in this survey). **Remarks**: The larva constructs an upper surface lobed blotch mine. The solitary larva produces scattered frass away from the mine perimeter. Pupation occurs within its mine in a 5.0 mm circular ridged pupal chamber. This latter observation differs from Braun (1908) but agrees with Maier and Davis (1989). This is a new host record for *Pediobius ocellatus*.

**Cameraria fletcherella** (Braun) (Fig. 64, 65). **Specimen data**: Lily Pond 46°50.892N 87°49.783W, 3 pupating larvae 16 Sep 2005, 1♂ after wintering, Lot 1649. **Distribution**: **CANADA**: QC; **U.S.A.**: CT, IL, KY, ME, NY, TX (De Prins and De Prins 2005). **Larval hosts**: *Quercus obtusifolia* Michx., *Q. tinctoria* Bartram, *Q. imbricaria* Michx., *Q. macrocarpa*; *Q. ilex*; *Q. rubra*; *Q. velutina* Lam., *Castanea dentata* (Marshall) Borkh. (De Prins and De Prins 2005). **Quercus rubra** (identified in this survey). **Parasitoids**: **Braconidae**: *Colastes (Shawiana) metallica* (Yu et al. 2012). **Remarks**: The larva is a solitary feeder. Frass is brown spotting away from the mine perimeter. The completed mine is a lobed upper surface blotch with two parallel wrinkles on its upper surface above its pupal chamber.

**Cameraria lentella** (Braun) (Fig. 66, 67). **Specimen data**: Borrow Pit 46°53.140N 87°56.919W, 21 larvae 25 Sep 2002, 1♂, 3♀ after wintering, Lot 1440. **Distribution**: **U.S.A.**: AZ, CT, GA, IL, ME, MD, NJ, NY, OH, VT (De Prins and De Prins 2005). **Larval hosts**: *Ostrya virginiana* (De Prins and De Prins 2005). **Ostrya virginiana** (identified in this survey). **Parasitoids**: None reported (Yu et al. 2012, Noyes 2018).

**Braconidae**: *Pholetesor sp. 1*, *Pholetesor sp. 2*, *Pholetesor sp. 3*; **Eulophidae**: *Chrysocharis occidentalis* (identified in this survey). **Remarks**: This gregarious species forms an upper surface irregular blotch mine frequently centered over the main vein. When completed, longitudinal...
Figures 58–65. Fig. 58. Cameraria aceriella: cocoon visible in completed mine. Fig. 59. Cameraria bethunella: adult. Fig. 60. Cameraria bethunella: completed mine with three wrinkles visible. Fig. 61. Cameraria betulivora: adult. Fig. 62. Cameraria corylisella: adult. Fig. 63. Cameraria corylisella: completed mine with cocoon. Fig. 64. Cameraria fletcherella: adult. Fig. 65. Cameraria fletcherella: completed mine with two wrinkles visible.
wrinkles are apparent on the upper mine surface. Frass appears as brown spotting on the mine floor. Mines were recovered in both mid-July and again in mid- to late September and both produced adults, confirming two generations at this location as Maier and Davis (1989) reported for Connecticut. The specimen of Phoeetesor recorded in this research are the first species of Braconidae reported as parasitic on C. lentella. This is a new host record for Chrysosochis orientalis.

*Camelaria ostryarella* (Chambers) (Fig. 68, 69). *Specimen data*: Borrow Pit 46°53.140N 87°56.071, 1 larva 25 Sep 2002, 1♂ after wintering, Lot 1988. Distribution: CANADA: QC; U.S.A.: CT, IL, NY, VT, De Prins and De Prins 2006.

*Larval hosts*: *Carpinus caroliniana* (De Prins and De Prins 2005).


*Deutizas syringella* (Fabricius) (Fig. 72). *Specimen data*: Stone House 46°50.627N 87°51.300W, 7 mines with multiple larvae in each 29 Jun 2000, 16♂, 1♀ 17-29 Jul 2000, Lot 1023; same site, 1 mine with nine larvae 20 Aug 2004, 1♂, 1♀ 14-16 Sep 2004, Lot 1524. Distribution: Palearctic; CANADA: NS, QC; U.S.A.: ME, MI, VT (De Prins and De Prins 2005).

*Sympiesis sericeicornis*. 

Gracillaria syringella (Fabricius) (Fig. 72). *Specimen data*: Stone House 46°50.627N 87°51.300W, 7 mines with multiple larvae in each 29 Jun 2000, 16♂, 1♀ 17-29 Jul 2000, Lot 1023; same site, 1 mine with nine larvae 20 Aug 2004, 1♂, 1♀ after wintering, Lot 1524. Distribution: Palearctic; CANADA: NS, QC; U.S.A.: ME, MI, VT (De Prins and De Prins 2005).

*Deutizas syringella* (Fabricius) (Fig. 72). *Specimen data*: Stone House 46°50.627N 87°51.300W, 7 mines with multiple larvae in each 29 Jun 2000, 16♂, 1♀ 17-29 Jul 2000, Lot 1023; same site, 1 mine with nine larvae 20 Aug 2004, 1♂, 1♀ after wintering, Lot 1524. Distribution: Palearctic; CANADA: NS, QC; U.S.A.: ME, MI, VT (De Prins and De Prins 2005).
Figures 66–73.

Fig. 66. *Cameraria lentella*: adult.

Fig. 67. *Cameraria lentella*: four completed mines with tentiform pupal areas visible.

Fig. 68. *Cameraria ostryarella*: adult.

Fig. 69. *Cameraria ostryarella*: two miners with one circular cocoon in *Ostrya virginiana*.

Fig. 70. *Cameraria saccharella*: adult.

Fig. 71. *Cameraria saccharella*: larva on vein and second larva perpendicular on same vein with some frass placed over veins.

Fig. 72. *Gracillaria syringella*: adult.

Fig. 73. *Leucanthiza dirce*: adult.
This species clearly has two generations per year at this survey site and agrees with Murdoch's (1967) observations.

*Leucanthisa direcilla* Braun (Fig. 73, 74). **Specimen data:** Ives Lake Rd. 46°50.475N 87°50.581W, 11 mines 27 Sep 2001, 0 adults, Lot 1256; same site, 1 larva 17 Sep 2006, Lot 1817. **Distribution:** CANADA: QC; U.S.A.: CA, KY, ME, MI, OH, VT (De Prins and De Prins 2005). **Larval hosts:** Canadensis borealis (L.), *Robinia* pseudoacacia, *R. viscosa* (Mason), and *Pnigalio inunctus* (Nees), and *Pnigalio elegantissimus* (Nees), *P. maculipes* (Howard), and *Pnigalio albipes* (identified in this survey). **Parasitoids:** **Eulophidae:** *Chrysoscharis occidentalis* (Howard, *Closterocerus trifasciatus*, *Pnigalio sp.*, *P. maculipes*, *P. uroplatae* (Howard), *Sympiesis trifasciatus*, *Pnigalio maculipes*, *C. viticola* (Ronan), and *Pnigalio sp.* (identified in this survey). **Remarks:** The mine is an irregular blotch on the upper leaf surface. There are one or three mines per leaf but only a single larva per mine. Dark frass particles are distributed in indistinct trails away from the mine perimeter. The larva is pale green upon exiting the upper leaf surface. It pupates in an oval white cocoon approximately 2.0 x 4.0 mm. These observations agree with those described by Petrice et al. (2000) for another northern Michigan population. This is a new host record for *Pediobius albigipes*.

*Macroscacbus robiniella* (Clemens) (Fig. 75, 76). **Specimen data:** Ives Lake Road 46°50.475N 87°50.581W, 11 mines 27 Sep 2001, 0 adults, Lot 1256; same site, 12 mines 24 Sep 2002; 1 larva 17 Aug 2004, 13 larvae 16 Sep 2006, Lot 1757; same site, 1 larva 30 Jun 2002, Lot 1350; same site, 12 larvae 16 Sep 2006, 4♀, 30 Sep – 04 Oct 2006, Lot 1757; same site, 1 larva 30 Jun 2007, 0 adult, Lot 1300; same site, 3 larvae 24 Sep 2008, 3♀, 12–13 Oct 2008, Lot 1903; same site, 1 cocoon 27 Aug 2009, 1♂, 13 Nov 2009, Lot 1942; Conway Bay 46°53.101N 87°48.706W, 9 larvae 25 Jun 2002, 3♂, 06 Jul 2002, Lot 1295; same site, 4 larvae 23 Sep 2002, 1♂, 08-12 Oct 2002, Lot 1347; Picnic Point 46°53.056N 87°50.590W, 12 larvae 05 Jun 2006, 3♀, 1♂ 18-22 Jun 2006, Lot 1698. **Distribution:** CANADA: BC, NL, NB, NS, YT; U.S.A.: CA, CO, CT, IL, KY, NV, UT (Kirichenko et al. 2016). **Larval hosts:** *Vicia caroliniana* Walt., *Mellotus sp.*, *Mellotis officinalis* (L.) Lam. (De Prins and De Prins 2005); *Lathyrus japonicus*, *Mellotis albuis Medik., Vicia caroliniana, Lupinus sp.*, Caragana sp. (Kirichenko et al. 2016). **Larvuidus japonicus** (identified in this survey). **Parasitoids:** *Brachonidae: Pholetesor bedelliae, Pholetesor saltaicus* (Mason), Pholetesor variabilis Whitfield (Yu et al. 2012). **Ichneumonidae:** *Diaglyptidea sp.* (identified in this survey). **Remarks:** There is usually only one mine per leaflet though occasionally two occur. This upper surface mine is a lobed blotch. Frass is expelled via a hole at the mine base, which is frequently pink. The yellowish larva exits the lower leaf surface to form an oval pale yellow, or rarely white, cocoon. Occasionally, larvae pupate in their mines with adults emerging on the upper leaflet surface. From recovery and emergence dates, there are two generations per year at HMC. This agrees with Kirichenko et al. (2016). The specimens of *Diaglyptidea* reared in this research are the first ichneumonids reported as parasitoids of *M. occulta*.

*Parectopa* sp. prob. *robiniella* Clemens (Fig. 79). **Specimen data:** Ives Lake Road 46°50.475N 87°50.581W, 2 larvae 17 Aug 2004, 0 adults, Lot 1517. **Distribution:**
Figures 74–81. Fig. 74. *Leucanthiza diricella*: vacated mine. Fig. 75. *Macrosaccus robiniella*: adult. Fig. 76. *Macrosaccus robiniella*: lower surface mine with pupal skin extended. Fig. 77. *Mirurapteryx occulta*: adult. Fig. 78. *Mirurapteryx occulta*: vacated mine. Fig. 79. Parectopa sp. prob. *robiniella*: early mine. Fig. 80. *Parornix conspicuella*: adult. Fig. 81. *Phyllocoenitis populiella*: adult.
Moench. (De Prins and De Prins 2005).

*Robinia pseudoacacia* 

*RLactia volubilis* (De Prins and De Prins 2005).


*Biota nigra* *L.*, *Eulophidae:* *Pholetesor circumscriptus,* *Pholetesor ornigis* (Yu et al. 2012).

**Remarks:** This easily recognized mine is a narrow sinuous trail on the upper leaf surface just below the epidermis. It is usually initiated and terminates near the leaf edge. Frass is a continuous central trail. When feeding is complete, the larva folds a small part of the leaf perimeter and then pupates within it in a white cocoon.

**Phyllobryctis alnologia** (Walsingham) (Fig. 83, 84, 85). **Specimen data:** Flat Rock 46°54.520N 87°55.355W, 3 mines 22 Sep 2011, 1♀ after wintering. Lot 2034.

**Distribution:** U.S.A.: CA, ME (De Prins and De Prins 2005). **Parasitoids:** *Ichneumonidae:* *Anisacris pampylina* (L.) (Yu et al. 2012). **Remarks:** The lower surface mine is an oval blotch confined by adjacent lateral veins. Feeding progresses upward at the mine perimeter then continuing inward. When fully fed, much of the upper leaf around the mine perimeter and toward its center is consumed. Larvae mining the upper sides of leaves of *A. incana,* as observed initially by Walsingham (1889) and repeated by others (Braun 1908, De Prins and De Prins 2005), was not observed in this rearing. That observation may have arisen when Walsingham observed larvae nearing completion of their feeding. The larval feeding pattern reported here is typical of species in this genus. Both mine surfaces of the feeding larva are illustrated. Pupation occurs in the mine with adult emergence from the lower leaf surface, which is also typical of many Phyllobryctis species.

**Phyllonorycter apparella** (Her- rich-Schäffer) (Fig. 86).

**Specimen data:** Jensen homestead 46°53.656N 87°53.131W, 7 mines 16 Aug 2004, 1♀ after wintering. Lot 1513. **Distribution:** U.S.A.: AL, BC, MB, NS, ON, QC, SK, KT; U.S.A.: AK, CO, CT, ME, MD, MA, MI, MN, NM, OH, VT, VA, WA (De Prins and De Prins 2005). **Parasitoids:** *Ichneumonidae:* *Eulophidae:* *Pediobius albipes* (identified in this survey).
Figures 82–89. Fig. 82. Phyllocnistis populiella: completed mine in *Populus tremuloides* with pupal leaf edge fold. Fig. 83. Phyllonorycter alnicolella: adult. Fig. 84. Phyllonorycter alnicolella: upper mine surface. Fig. 85. Phyllonorycter alnicolella: lower mine surface. Fig. 86. Phyllonorycter apparella: adult. Fig. 87. Phyllonorycter auronitens: adult. Fig. 88. Phyllonorycter basistrigella: adult. Fig. 89. Phyllonorycter basistrigella: lower mine surface with cocoon.
Braconidae: Pholetesor sp. 3 and Pholetesor (♀); Eulophidae: Clostercerus trifasciatus (identified in this survey). Remarks: The completed mine of this solitary miner is the usual lower surface oval tentiform shape. Also, the larva pupates in its mine as is usual for the genus. This is a new host record for Clostercerus trifasciatus.


Phyllonorycter lucetiella (Clemens) (Fig. 94, 95). Specimen data: BMC entrance gate 46°55.926N 87°50.012W, 9 larvae 27 Sep 2000, 1♂, 1♀ after wintering, Lot 1097; Borrow Pit at Howe Lake 46°53.140N 87°56.919W, 9 larvae 20 Sep 2003, 1♂, 2♀ after wintering. Lot 1445. Distribution: CANADA: QC, U.S.A.: CT, IL, KY, ME, MI, NY OH.
Figures 90–97. Fig. 90. Phyllonorycter clemensella: adult. Fig. 91. Phyllonorycter clemensella: lower surface of completed mine. Fig. 92. Phyllonorycter ledella: adult. Fig. 93. Phyllonorycter ledella: completed mine (reared from Upper Peninsula but not at HMC). Fig. 94. Phyllonorycter lucetiella: adult. Fig. 95. Phyllonorycter lucetiella: lower surface of completed mine with frass around its perimeter. Fig. 96. Phyllonorycter lucidicostella: adult. Fig. 97. Phyllonorycter lucidicostella: lower surface of completed mine.
Lot 1437. Quarry-Fen 46°53.839N 87°53.607W, 4 24 Sep 2002, 3♀ after wintering, Lot 1352; 46°54.520N 87°55.355W, 5 completed mines (Fig. 96, 97).

Perimeter. Pupation occurs in the mine with frass tightly appressed to the mine perimeter. Pupation occurs in the mine with its oval cocoon clearly visible. This is a new host record for Ageniaspis bicoloripes.

**Phyllonorycter lucidicostella** (Clemens) (Fig. 96, 97). **Specimen data:** Flat Rock 46°54.520N 87°53.607W, 5 completed mines 24 Sep 2002, 3♂ after wintering, Lot 1352; Quarry-Fen 46°53.839N 87°53.607W, 4 larvae 17 Sep 2003, 1♂, 1♀ after wintering, Lot 1437. **Distribution:** CANADA: ON, QC; U.S.A.: AL, CT, IL, KY, ME, MI, NY, PA, VT (De Prins and De Prins 2005). **Larval hosts:** Acer saccharinum, A. floridanum (Chapm.) Pax., A. saccharum (Freeman) (Fig. 96).

**Remarks:** The completed lower surface mine is confined by adjacent veins with frass tightly appressed to the mine perimeter. Pupation occurs in the mine with its oval cocoon clearly visible. This is a new host record for Ageniaspis bicoloripes.

**Phyllonorycter martiella** (Traher) (Fig. 98, 99). **Specimen data:** Quarry-Fen 46°53.839N 87°53.607W, 3 larvae, 17 Sep 2003, 1♂ after wintering, Lot 1439. **Distribution:** CANADA: BC, NS, QC; U.S.A.: KY, ME, MI, NC, VT (De Prins and De Prins 2005). **Larval hosts:** Betula lenta, Betula sp. (De Prins and De Prins 2005); B. papyrifera (identified in this survey). **Parasitoids:** None reported (Yu et al. 2012, Noyes 2018). **Remarks:** The completed lower surface mine is confined by adjacent veins. Frass is scattered throughout the mine. Pupation occurs in an oval cocoon at the mine center. The specimen of Pholetesor reared in this research is the first species of Braconidae reported as parasitic on *P. martiella*.

**Phyllonorycter nipigon** (Freeman) (Fig. 100). **Specimen data:** Jensen Homestead 46°53.656N 87°53.131W, 4 completed mines 16 Aug 2004, 1♀, 2♂ Aug 2004, Lot 1512. **Distribution:** CANADA: AB, BC, ON, QC, SK; U.S.A.: AK, CA, CO, ID, MI, MN, MT, WA, WY (De Prins and De Prins 2005). **Larval hosts:** Populus angustifolia James, *P. balsamifera, P. balsamifera trichocarpa* (Torr. and Gray), *P. × acuminata Rhdyb. (P. angustifolia × deltoides Marsh.), P. fremontii* S. Watson, *P. nigra, P. tremuloides* (Davis and Deschka 2001, De Prins and De Prins 2005); *P. balsamifera* (identified in this survey). **Parasitoids:** Eulophidae: Sympiesis gordius, *S. sericeicornis, S. stigma.* Zagrommoma americanum Girault, *Z. multilineatum* (Ashmead) (Noyes 2018). **Eulophidae:** Closterocephus trifasciatus (identified in this survey). **Remarks:** The completed mine of this solitary larva is an elongate lower surface tentiform blotch. Frass consists of black particles at the mine perimeter. This is a new host record for Closterocephus trifasciatus.

**Phyllonorycter ostryaeofoliola** (Clemens) (Fig. 101). **Specimen data:** Mt. Lake Boat House 46°56.720N 87°50.333W, 26 larvae 28 Sep 2000, 2♂, 3♀ after wintering, Lot 1099; trail end to Cedar Creek 46°58.279N 87°49.114W, 11 larvae 25 Sep 2001, 1♂, 1♀ after wintering, Lot 1244. **Distribution:** CANADA: NS, ON, QC; U.S.A.: CT, IL, KY, ME, NY, VT (De Prins and De Prins 2005). **Larval hosts:** Ostrya virginiana (De Prins and De Prins 2005); *O. virginiana* (identified in this survey). **Parasitoids:** Eulophidae: Achrysocharoides arienascapus (Miller) (Noyes 2018); *Bracconidae: Pholetesor ornigis, Rhyssopolis decorator* (Haliday) (Yu et al. 2012). **Eulophidae:** Chrysocharis sp.; *Bracconidae: Rhyssopolis decorator* (identified in this survey). **Remarks:** One to five mines occur per leaf though the larva is solitary. The mine is typical of several species in this genus being lower surface and tentiform. Frass is generally scattered through the mine though occasionally clumped. This is a new host record for Chrysocharis sp.

**Phyllonorycter salicifoliola** (Chambers) (Fig. 102, 103). **Specimen data:** Stone House 46°50.627N 87°51.300W, 6 mines 17 Aug 2004, 3♂, 3♀ 28 Aug - 01 Sep 2004, Lot 1516. **Distribution:** CANADA: AB, BC, ON, QC; U.S.A.: CA, CO, CT, ID, IL, KY, ME, MD, MA, MI, NJ, NY, OH, VA, WA (De Prins and De Prins 2005). **Larval hosts:** Populus balsamifera, *P. grandiflora* (Cham -
Figures 98–105. Fig. 98. *Phyllonorycter martiella*: adult. Fig. 99. *Phyllonorycter martiella*: completed and vacated mine. Fig. 100. *Phyllonorycter nipigon*: adult. Fig. 101. *Phyllonorycter ostryaefoliella*: adult. Fig. 102. *Phyllonorycter salicifoliella*: adult. Fig. 103. *Phyllonorycter salicifoliella*: lower surface of completed mine. Fig. 104. *Protolithocolletis lathyri*: adult. Fig. 105. *Protolithocolletis lathyri*: lower leaf surface with mined leaf edge rolled under.


Braconidae: Pholetes sp. 1 (identified in this survey). Remarks: This species forms a typical Phyllonorycter oval lower surface tentiform mine. Frass is scattered around the terminal mine and balled near the mine apex. Pupation occurs within the mine. A single wrinkle, parallel with the long axis of the mine, is evident on the lower leaf surface.


Braconidae: Pholetes salicifoliiatae, Rhysipolis pallipes (Provancher) (Yu et al. 2012). Eulophidae: Sympiesis sericeicornis (identified in this survey). Remarks: The completed lower surface mine is a long oval tentiform shape and placed on one side of the main vein. Wrinkling of the lower mine surface causes the leaflet to roll downward partially covering the mine. Feeding frequently extends to the upper surface appearing as stippling and at times with complete removal of all green tissue. The larva pupates inside the mine. This is its first published location in Michigan but its second recovery. An adult specimen in the NMNH was recovered by A. Braun with label reading, “30 Jul 1943, Huron Mountains, MI.” No larval host data were mentioned. Combining Braun’s adult recovery date with emergence dates reported here, there appears to be at least two generations per year in this survey area. This is a new host record for Sympiesis sericeicornis.

Nepticulidae

Glaucopis saccharella (Braun) (Fig 106, 107). Specimen data: Loop Road 46°50.627N 87°51.300W, 1 larva 13 Sep 2005, 1♂ after wintering, Lot 1652. Distribution: CANADA: ON, QC; U.S.A.: OH (Wilkinson and Scoble 1979). Larval hosts: Acer rubrum, A. saccharum (Braun 1917); Quercus rubra (Wilkinson and Scoble 1979); A. saccharum (identified in this survey). Parasitoids: None reported (Yu et al. 2012, Noyes 2018). Remarks: The very long linear upper surface mine gradually widens along its entire length. Frass is deposited as a continuous central line of black particles along the entire mine. The terminus is approximately 1.3 mm wide. The pale yellow-green larva exits the upper surface prior to pupating in a flat brown 2.1 × 1.2 mm oval cocoon. A previous rearing, Lot 1435 recovered in 2003, has a similar mining pattern, but the adult did not agree in coloring with the adult description of Braun (1912).

Stigmella sp. prob. macrocarpae (Freeman) (Fig. 108). Specimen data: Lily Pond, 46°50.889N 87°49.783W, 4 larva (2 dead) 18 Sep 2006, 0 adult, Lot 1761; same site, 1 larva 18 Sep 2006, 0 adults, Lot 1771. Distribution: CANADA: BC, ON; U.S.A.: AR, FL, IL, KY, MA, MI, NY, OH, PA, VA (Newton and Wilkinson 1982). Larval hosts: “Red and scarlet oaks, and probably Chestnut” (Braun 1917); Castanea dentata, Quercus alba, Q. macrocarpa, Q. palustris Münch., Q. rubra (Wilkinson and Scoble 1979). Quercus rubra (identified in this survey). Parasitoids: None reported (Yu et al. 2012, Noyes 2018). Braconidae: Gnaptopodon nepticulae (Rohwer) (identified in this survey). Remarks: The upper surface linear mine begins at a vein, gradually widening during its development. Frass is deposited across the mine through most of its length. At approximately mid-length frass particles are arranged in concave bands across the mine. In the last few millimeters of deposition the frass bands are narrowed covering less than a third of the central mine width. The terminal mine width is 1.8–2.0 mm. The full grown larva is bright green, exits the lower leaf surface, and forms a white cocoon approximately 1.3 × 2.5 mm. These observations agree especially well for this species as described by Braun (1917) but also of those provided by Lindquist and Harnden (1970) and Wilkinson and Scoble (1979). Gnaptopodon nepticulae would be the first species of Braconidae reported as parasitic.
Figures 106–112. Fig. 106. *Glaucoplepis saccharella*: adult. Fig. 107. *Glaucoplepis saccharella*: completed and vacated mine. Fig. 108. *Stigmella* sp. prob. *macrocarpae*: completed and recently vacated mine. Fig. 109. *Stigmella quercipulchella*: adult. Fig. 110. *Stigmella quercipulchella*: completed and vacated mine. Fig. 111. *Coptotriche citrinipennella*: adult. Fig. 112. *Coptotriche citrinipennella*: completed mine in *Quercus imbricaria*. Mine from Lower Peninsula.
on S. macrocarpae pending unequivocal host identification.

Stigmella quercipulchella (Chambers) (Fig.109, 110). Specimen data: Lily Pond 46°50.892N 87°49.783W, 2 larvae 29 Sep 2000, 1♀ after wintering, Lot 1100. Distribution: Canada: ON; USA: IL, KY, OH, PA (Newton and Wilkinson 1982). Larval hosts: Quercus rubra (Wilkinson and Scoble 1979); Q. palustris, Q. marilandica Münchhausen (Newton and Wilkinson 1982); Quercus rubra (identified in this survey). Parasitoids: None reported (Yu et al. 2012, Noyes 2018). Remarks: The upper surface mine begins at a secondary vein. It meanders and widens gradually the first half of its length. The last half is significantly wider measuring 4.5 mm across at its terminus. Frass is distributed in a thin central line throughout the mine length. The single pale green larva exits the upper leaf surface. It pupates in a tan cocoon 2.3 × 1.7 mm.

Tischeriidae

Coptotriche citrinipennella (Clemens) (Fig. 111, 112). Specimen data: Breakfast Roll Mountain 46°51.015N 87°49.601W, 1 larva 26 Sep 2000, 1♀ after wintering, Lot 1095. Distribution: Canada: NS, ON, QC; USA: AR, DC, IN, KY, MA, MO, NJ, NY, NC, OH, PA, TX, VA, WV (Braun 1972); Mi (Nielsen 1998). Larval hosts: Quercus imbricaria, Castanea dentata (Braun 1972); Q. rubra (identified in this survey). Parasitoids: Braconidae: Pholetesor ornignis (Yu et al. 2012). Eulophidae: Chrysoscharis sp., Pnigalio sp. (Noyes 2018). Remarks: The mature upper surface mine is elongate and constructed along the leaf edge. As the mine enlarges, the leaf edge is rolled onto the upper surface hiding most of the mine. The larva pupates in its mine, emerging from the distal end. Though this was the only specimen observed during the survey, RJP has found it the most common species mining Quercus in both Michigan Peninsulas. This is a new record for the genera Chrysoscharis and Pnigalio.

Acknowledgments

The first author is most thankful to David Gosling, former Huron Mountain Wildlife Foundation (HMWF) Director, for approving the initial proposal to survey leaf mining insects, guidance to various habitats, and encouragement to continue surveying even when recoveries were at first unexpectedly few. Kerry Woods (Bennington College, Vermont), current HMWF Director, is also sincerely thanked for his continued support and patience with this work. RJP is also most grateful to the Huron Mountain Wildlife Foundation for its years of financial support for field work and providing housing while on site. RJP is also most indebted to William and Anne Manierre (both recently deceased) for sharing their lifelong knowledge of HMC sites, guidance to a variety of them, personal friendship and encouragement, and their most generous hospitality during my years of visits. Additionally, RJP very much appreciated Fred Rydholm’s (deceased) guidance over lands and his eagerness to share the history of the HMC, its world renowned visitors, and his friendship. RJP also thanks Wayne Thorpe, HMWF manager, for his historical knowledge and assistance arranging accommodations at Stonehouse, the research facility. To Frederick W. Stehr (Emeritus curator, MSUC), RJP is indebted for initially inviting him to work at the A.J. Cook Arthropod Research Collection at Michigan State University (MSU) as an Adjunct Curator. Indebtedness is also owed to Anthony Cognato, current Collection Director for his continued personal and financial support for visits to the National Museum of Natural History to identify specimens and for reviewing an earlier draft of this manuscript. Continued work in the collection would not have been possible without the assistance of Gary Parsons, Collection Manager, and his continued encouragement in my effort to focus on leafminers.

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Michigan State University, was most helpful in creating the general map of HMC in upper Michigan. We thank Gary Parsons and Bernice DeMarco for their reviews of an earlier draft of this manuscript and offering helpful improvements. We also thank two anonymous reviewers who offered very helpful suggestions and significantly improved the later draft. RJP also acknowledges indebtedness to Eugene Rhodes Thompson (deceased) for his insightful question as this project began, “Are you taking pictures?” Finally, RJP thanks his wife, Helen, for her patience during many absences, recording emergence dates, assisting on several field trips, and her constant support throughout these years of survey.

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**Literature Cited**


Ciidae of Michigan (Insecta: Coleoptera)

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Abstract

The family Ciidae Leach, 1819, (Insecta: Coleoptera) occurs worldwide with approximately 720 species. In the United States there are 84 species in 13 genera. Given their relatively small size (~0.5 to 6 mm) and cryptic habitats, feeding in decaying fungi, recent regional fauna studies are lacking including the northeastern United States. To alleviate this gap in knowledge, in part, we review and identify 2,123 undetermined specimens collected in Michigan. We provide new state records for four species: Ceracis pecki Lawrence, 1971, Cis americanus Mannerheim, 1852, Cis submicans Abeille de Perrin, 1874, Dolococis manitoba Dury, 1919, which increases the total for Michigan to 25 species and update records for Michigan counties. In addition, we provide a modified key to Michigan species.

Keywords: Dichotomous key, horned beetles, faunistic study

Material and Methods

Approximately 2,123 undetermined specimens were examined and identified to species using the dichotomous key found in Lawrence (1971). The MSUC specimens were databased at http://symbiota4.acis.ufl.edu/scan/portal/index.php. General distribution for each species was determined using Lawrence (1971) except for the distribution of Ceracis (Lawrence 1967). Specimens were borrowed or used from the following entomological collections.

Field Museum of Natural History (FMNH)
Michigan State University Collection (MSUC)
University of Michigan Museum of Zoology (UMMZ)
Wisconsin Insect Research Collection (WIRC)

Results and Discussion

Previously, there were only ten species reported from Michigan (Lawrence
Table 1: All Ciidae species known to occur in Michigan. Source is the publication that lists the state record or a new record discovered in this study.

<table>
<thead>
<tr>
<th>Species</th>
<th>Author</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ceracis minutissimus</td>
<td>(Mellié, 1849)</td>
<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Ceracis pecki</td>
<td>Lawrence, 1971</td>
<td>New State Record</td>
</tr>
<tr>
<td>Ceracis punctulatus</td>
<td>Casey, 1898</td>
<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Ceracis singularis</td>
<td>(Dury, 1917)</td>
<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Ceracis sallei</td>
<td>(Mellié, 1849)</td>
<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Ceracis thoracicornis</td>
<td>(Ziegler, 1845)</td>
<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Cis americanus</td>
<td>Mannerheim, 1852</td>
<td>New State Record</td>
</tr>
<tr>
<td>Cis castlei</td>
<td>(Dury, 1917)</td>
<td>Downie and Arnett, 1996</td>
</tr>
<tr>
<td>Cis fuscipes</td>
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<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Cis horridulus</td>
<td>Casey, 1898</td>
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<td>Cis levettei</td>
<td>(Casey, 1898)</td>
<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Cis striatulus</td>
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<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Cis submicans</td>
<td>Abeille de Perrin, 1874</td>
<td>New State Record</td>
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<td>Cis subtilis</td>
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<tr>
<td>Cis tristis</td>
<td>Mellié, 1849</td>
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</tr>
<tr>
<td>Dolococis manitoba</td>
<td>Dury, 1919</td>
<td>New State Record</td>
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<tr>
<td>Hadreule blaisdelli</td>
<td>(Casey, 1900)</td>
<td>Lawrence, 1982</td>
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<td>Octotemnus glabriculus</td>
<td>(Gyllenhal, 1827)</td>
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<td>Sulcacis lengi</td>
<td>Dury, 1917</td>
<td>Lawrence, 1982</td>
</tr>
<tr>
<td>Sulcacis curtulus</td>
<td>(Casey, 1898)</td>
<td>Lawrence, 1982</td>
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</tbody>
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1967, Lawrence 1971). Subsequent records (Lawrence 1982, Downie and Arnett 1996) and our new state records for Ceracis pecki Lawrence, Cis americanus Mannerheim, Cis submicans Abeille de Perrin, and Dolococis manitoba Dury increase the known Michigan fauna to 25 species. Ceracis Mellié and Cis Latreille species mostly represent this fauna (Table 1). The most abundant species found among the Michigan specimens were *Cis fuscipes* Mellié, *Cis levettei* (Casey) and *Ceracis thoracicornis* (Ziegler). The previous studies that catalogued Michigan species did not give county records (Lawrence 1982, Thayer and Lawrence 2002). This study gives updated county records for *Ceracis minutissimus* (Mellié), *Ceracis singularis* (Dury), *Ceracis thoracicornis*, *Cis castlei* (Dury), *Cis fuscipes* Mellié, *Cis fuscipes*, *Cis levettei* Casey and *Ceracis thoracicornis* Mellié, and *Ceracis pecki* Lawrence, *Ceracis singularis* (Dury), *Ceracis thoracicornis* (Ziegler), *Cis creberrimus* Mellié, *Cis fuscipes* Mellié, *Cis levettei* Mellié, and *Ceracis thoracicornis* (Ziegler) Casey.

New Records (*new record*)


*Cis americanus* Mannerheim, 1852 Michigan, Gogebic County, Wakefield. 20 Aug 1964. W. Suter Coll. (FMNH-2).


*Cis submicans* Abeille de Perrin, 1874 Michigan, Clare Co. 07 May 1938. R. R. Dreisbach coll. (MSUC-2).


Systematics

The keys provided below, modified from Lawrence (1971), apply only to genera and species of Ciidae occurring in Michigan. See Lopes-Andrade et al. (2016) for habitus images for many of the Michigan species.

**Key to the Ciidae genera**

1 Prosternal process short, not extending to middle of coxae; first abdominal ventrite of male with triangular flap concealing pubescent sex patch. Distal third of tibia bearing spines on the outer edge. Antennae 8-segmented........... *Octotemnus* Mellié Prosternal process extending beyond middle of coxae; first abdominal ventrite may have a sex patch in male, but never bearing flap-cover. Tibiae and antennae variable.................................................................2

2 Prosternum in front of coxae almost twice as long as prosternal process ...............................................................

.............................................................................................................. *Hadreule* C. G. Thomson Prosternum in front of coxae not or only slightly longer than prosternal process .................................................................................................................................................................................3

3 Outer angle of protibia expanded, rounded, and bearing several spines..............4 Outer angle of protibia not as above, blunt and angulate, not bearing spines.................................................................................................................................................................................................4

4. Lateral edges of pronotum visible from above for their entire lengths; anterior pronotal angles slightly produced .............................................. *Strigocis* Dury Lateral edges of pronotum not visible from above for their entire lengths; anterior pronotal angles not produced .................................................................................................................................................................5

5. Prosternal process thin; short setae on pronotum and elytra, usually not visible under microscope, head and apex of pronotum in males usually produced to form horns.................................................................................................................. *Ceracis* Mellié Prosternal process wide; setae visible under scope, pronotum never produced .................................................................................................................................................................................6

6. Lateral edges of pronotum slightly produced; pronotum not produced to form horns......................................................................................... *Sulcacis* Dury

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6. Outer apical angle of protibia narrowly rounded; elytral punctation single and uniform; body elongate and parallel-sided. ...................... Orthocis Casey

Outer apical angle of protibia mostly produced and dentate or blunt and angulate; elytral punctation not uniform ........................................ 7

7. Antenna with 10 antennomeres ........................................... Cis Latreille

Antenna with 9 antennomeres ........................................... 8

8. Outer apical angle of protibia rounded; elongate, pronotum never produced ................................................................. Dolichocisi Dury

Outer angle of protibia produced and dentate; prosternal process thin; pronotum in males produced ........................................ Plesiocis Casey

**Key to the genus: Sulcacis**

There are two North American species, both are found in Michigan.

1. Antennae 9-segmented .......................................................... S. lengi

Antennal 10-segmented ......................................................... S. curtulus

**Key to the genus: Orthocis**

There are five North American species and only *O. punctatus* is found in Michigan (Lawrence, 1982).

**Genus: Dolichocis**

There are two North American species and one of them is found in Michigan. *Dolichocis manitoba* has been reported from the Northern regions of North America.

**Genus: Strigocis**

There are three North American species and two of them are found in Michigan (Lawrence, 1982). *Strigocis opalescens* (Casey, 1898) has been reported from Wayne County, Michigan (Lawrence 1971). *Strigocis opacicollis* Dury has also been reported from Michigan (Lawrence 1982).

1. Margin of pronotum with a wide, raised lip; elytral hairs colorless .......................................................... S. opalescens

Margin of pronotum narrow, now raised; elytral hairs yellow .......... S. opacicollis

**Genus: Hadreule**

*Hadreule blaisdelli* (Casey, 1900) has been reported from Allegan County, Michigan (Lawrence, 1971). This species has a more western distribution, but extends to eastern parts of the country.

**Genus: Plesiocis**

*Plesiocis cribrum* Casey, 1898 is the only species in this genus that is found in North America and has been reported from Iosco County, Michigan (Lawrence, 1971).

**Genus: Octotemnus**

*O. glabriculus* is the only North American species and it has a widespread distribution throughout North America (Lawrence 1971).

**Genus: Ceracis**

There are 20 North American species and six occur in Michigan. Males occurring in Michigan can be distinguished by the presence of a sex patch on the first abdominal ventrite and most of the species have tubercles on the frontoclypeal ridge or on the apex of the pronotum. This key has a combination of male and female characters, where applicable.
Key to the genus Ceracis

1. Antennae with 10 antennomeres .............................................. Cer. singularis
2. Antennae with less than 10 antennomeres .................................. 2
3. Antennae with 9 antennomeres ................................................ 3
4. Antennae with 8 or fewer antennomeres ..................................... 4

5. Elytral punctures usually smaller than those on pronotum and separated by more than one puncture diameter; males bearing two long, deeply emarginate tubercles on pronotum .............................................. Cer. thoracicornus

Pronotal punctures equal in size to elytral punctures and separated by less than one puncture diameter; males with two horns arising from pronotum, but not as long or as emarginate ............................................. Cer. pecki
6. Elytral punctation coarser and denser than that of pronotum ... Cer. minutissimus

Elytral punctation finer and sparser than or as fine and sparse as that of pronotum ..................................................... 6

7. Pronotal punctation about as fine and sparse as elytral punctation, the punctures usually separated by more than one puncture diameter; males with an ovoid sex patch on first abdominal ventrite ............................................. Cer. sallei (Mellié)

Pronotal punctation coarser and denser than elytral punctation, the punctures usually separated by less than one puncture diameter; male with a circular sex patch on first abdominal ventrite ............................................. Cer. punctulatus Casey

Key to the genus Cis

There are 43 North American species and ten are found in Michigan.

1. Body flattened or subcylindrical ............................................... 2
   Body not flattened, cylindrical ............................................. 3

2. Body flattened, male with fovea on first visible abdominal sternite; frontoclypeal ridge in male with 4 tubercles ............................................. C. creberrimus

Body subcylindrical, fovea not present on first visible abdominal sternite in male ................................................................. C. horridulus Casey

3. Setae minute, not or barely visible under microscope ............... C. levettei

Setae long, visible under microscope .................................. 4

4. Elytral punctation biordinal in size, with the smaller punctures bearing setae... 5

Elytral punctation not biordinal in size ..................................... 6

5. Lateral margin of pronotum widely expanded; in dorsal view, pronotal margins visible for the entire lengths from above ............................................. C. castiei

Lateral margins of pronotum not or barely expanded; in dorsal view, pronotal margins not visible for the entire lengths from above ............................................. 7

6. Elytral punctation serrate; lateral margins of pronotum weakly to not crenulate ................................................................. C. fuscipes

Elytral punctation confused, not distinctly serrate; lateral margins of pronotum coarsely and distinctively crenulate ................................. C. submicans

7. Prosternum carinate; elytral punctation distinctly of two sizes; in male, apex of pronotum bearing two subtriangular processes ......................... C. americanus

Prosternum not distinctly carinate; elytral punctation of two sizes, but indistinctly so; apex of pronotum in male always simple .................................................. 8

8. Elytral punctation not seriate; in male, frontoclypeal ridge bearing two tubercles; abdominal fovea absent ............................................. C. subtilis

Elytral punctation seriate ..................................................... 9

9. Pronotal punctation dense, less than half a diameter between punctures; elytral bristles colorless ............................................. C. tristis Mellié

Pronotal punctation sparse with more than half a diameter between punctures; elytral bristles yellow ............................................. C. striatulus
Acknowledgments

We thank these curators and/or managers for the loan of specimens or assistance: Crystal Meier (FMNH), Mark O’Brien (UMMZ), Gary Parsons (MSUC), Craig Brabant and Daniel Young (WIRC). The comments of two anonymous reviewers improved earlier drafts of this publication which we greatly appreciate.

Literature Cited


Materials and Methods

On 22 August 2015, a live adult female of the cicada Neotibicen linnei (Smith & Grossbeck) (Hemiptera: Cicadidae) was found clinging to a fence post at a residential complex near the village of Merrimac (43.3733°N, 89.6235°W), Sauk County, northwest of Madison, Wisconsin. The sex of a cicada is easily determined by the external genitalia. At the time, many males of this cicada species were calling from several large trees in the area. Several exuviae of this cicada species were found on nearby trees. As N. linnei is the only species of this genus occurring at this locality, it is assumed that the exuviae were those species. The unusually docile cicada was easily collected by hand and subsequently identified as N. linnei by matching it with several specimens in the collections of the Milwaukee Public Museum (from the western counties of Vernon and Grant) and from records for Sauk and Dane counties provided by Thomas E. Moore (pers. comm. 1970). It was suspected that the female N. linnei had been parasitized owing to how easily it was captured. Therefore, the cicada was kept in a zip-lock plastic bag to determine if any parasitoids would emerge.

Results

The cicada lived for five days (27 August) at the end of which five dipteran larvae were found crawling in the bag. Two days later, four reddish-brown puparia were formed, with one larva remaining. A fifth puparium was found five days later (1 September). The puparia were transferred to a plastic vial containing moistened tissue paper to prevent desiccation. The now-dead cicada was not examined for exit holes. The vial was kept in a non-heated garage over the winter and one adult dipteran eclosed on 19 June 2016, some ten months after pupation. Adults from the remaining puparia failed to emerge for unknown reasons. The dipteran specimen was sent to Drs. James O’Hara and Bradley Sinclair at the Canadian National Insect Collection (CNC) (Ottawa) for taxonomic identification. The specimen turned out to be a male Emblemasoma erro (Aldrich) (Diptera: Sarcophagidae) based upon barcoding and matching specimens in the CNC (Bradley Sinclair, pers. comm. March 2019). The specimen is housed in the CNC.

Discussion

This observation constitutes an unusual published record of E. erro parasitizing a female Neotibicen cicada. Although this observation and conclusion are very preliminary, it suggests a line of further research on the interaction of dipteran parasitoids and cicadas. Sarcophagids typically attack only singing male cicadas cuing in on acoustical signals (Soper et al. 1976). Perhaps an added caveat is that sarcophagids such as E. erro visually cue in on mute female cicadas in Wisconsin.
close proximity to singing males, thereby expanding parasitism to both sexes. There have been previous reports of female cicada parasitization by sarcophagids (e.g. Super et al. 1976; Stucky 2015). One mechanism is *E. erro* female following a female *Megatibicen dorsatus* (Say) in flight and larvipositing on it. This could be the case in the present instance. Thus, visual cues play a key role in successful larvipod larviposition on female cicadas. *Emblemasoma erro* is known to attack a male cicada in flight soon after calling (Stucky et al. 2015), unlike other *Emblemasoma* which attack stationary singing cicadas. The observation of *E. erro* attacking *N. linnei* is a new record as well. And while limited, this observation suggests a possible synchronization of the annual lifecycle of cicada sarcophagid parasitoids with the adult emergence cycles of their annual cicada hosts. While *E. erro* is widely distributed from Canada to Texas (Pape 1996), *N. linnei* is distributed across much of the eastern half of the United States (Sanborn and Phillips 2013), but not Texas. Therefore, the interaction of *E. erro* with female individuals warrants further study. While clearly very limited, this observation could be a basis for further study. Successful sarcophagid attacks on female cicadas could impact reproductive capacity.

Acknowledgments

My sincere thanks to Drs. James O’Hara and Bradley Sinclair (CNC) for their assistance and suggestions. Drs. Sinclair and T. E. Moore greatly improved an earlier draft of the manuscript. I thank the Milwaukee Public Museum for generous support of my status as Curator Emeritus of Zoology. Thanks, too, to Roberta Gordon for wonderful hospitality and support at Summer Oaks, Merrimac, Wisconsin and for introducing me to the study locality.

Literature Cited


New findings continue to reveal previously unrecorded genera and species of insects, chiefly native Coleoptera, but also those representing four separate orders and families, mainly from southern Wisconsin. While a majority of specimens have been taken at or near the author’s residence in the town of Oregon, Dane County, a single specimen of the introduced pentatomid, *Picromerus bidens* (Linnaeus, 1758), was captured in northern Wisconsin (Oconto County). Although different collecting strategies were employed, a majority of those reported were attracted to UV lights; others were found beneath the bark of logs/trees; one was found crawling on a rock; and one was reared from cut hickory limbs. These genera or species occur respectively within the following eleven families: Leiodidae, Monotomidae, Cucujidae, Cryptophagidae, Ciidae, Tetratomidae, Curculionidae, Pentatomidae, Glyphipterigidae, Phoridae, and Pteromalidae. All but one of these insects were collected at or near the author’s residence (Dane County); the pentatomid was taken in northern Wisconsin (Oconto County). Three of the four non-Coleoptera fauna are introduced species.

**Keywords:** insects, Wisconsin

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**Coleoptera**

**Leiodidae**

*Gelae parvulum* (LeConte, 1878). – On 27 May 2016, a single specimen of the round fungus beetle *G. parvulum* was collected at a UV light at the author’s residence (42° 54’14.75”, –89° 25’25.52”). Previously, this species was classified within the large genus *Agathidium* Panzer 1797 (Downey and Arnett 1996a: 333) but has since been transferred to its current generic assignment (Miller and Wheeler 2004). The principal characters of *Gelae* are antennae with 11 antennomers, a distinct 3-segmented club, no supraocular carina, no postocular temporum, 5-4-4 female tarsal formula, and obsolete dorsal punctation (Miller and Wheeler 2004). As therein defined, the genus *Gelae* contains eight species, two others of which, *G. parile* (Fall, 1934) and *G. cognatum* (Matthews, 1887), also were transferred from *Agathidium*. Five new species were also described (Miller and Wheeler 2004). *Gelae parvulum* was previously recognized from Michigan and California (Miller and Wheeler 2004). Beetles in these genera are highly contractile, capable of closing themselves up into a hemispherical ball. Leiodidae was formerly known to contain 47 species in the U.S. and Canada (Peck 2001). These beetles inhabit moist forest habitats, where they feed on slime molds under bark in decaying logs. No genital dissection was performed on the specimen.

**Monotomidae**

*Europs pallipennis* (LeConte, 1861). – A single specimen of the root-eating beetle *E. pallipennis* was taken at a UV light at the author’s residence on 13 June 2017. Within the genus *Europs* Wollaston, 1854, five species are identified from North America, whose distribution occurs mainly in the eastern, southeastern, and southwestern states (Bousquet 2002). *Europs pallipennis* is the only species found in the northeast, having
been reported from Connecticut, Florida, Indiana, Maryland, New York, Pennsylvania, and Virginia (Downey and Arnett 1996b: 988). It was formerly classified under the family Rhizophagidae, but has been transferred to a subfamily of Monotomidae. Beetles of this family live under tree bark, and most are believed to feed on fungus or fungal byproducts, although the larvae of some species (e.g., Rhizophagus Herbst, 1793) are predaceous (Bousquet 2002). The head and pronotum of *E. pallipennis* are reddish-brown to piceous, but the elytra are paler, with a rounded medial apical spot (Evans 2014: 282).

**Cucujidae**

*Pediacus subglaber* Leconte, 1854. – On 30 June 2017, a single specimen of the flat bark beetle *P. subglaber* was collected at a UV light at the author’s residence. Along with *Cucujus Fabricius, 1775, Pediacus Shuckard, 1839* is one of only two genera found in North America (Thomas 2002). Originally assigned species status, this beetle was subsequently synonymized with a subspecies of *P. depressus* (Herbst, 1797), before being revalidated by Thomas (2003). *Pediacus subglaber* has a primarily Appalachian distribution, being known from the following provinces and states: Canada: Ontario, Quebec; U.S.: Alabama, District of Columbia, Georgia, Illinois, Indiana, Kentucky, Maryland, Michigan, North Carolina, Pennsylvania, Tennessee, Vermont, Virginia, and West Virginia (Thomas 2003). It is the only species of the genus found in eastern North America south of New England (Thomas 2003). The head and pronotum are reddish brown, the latter bearing a crenulate margin, and the elytra are a paler yellowish color (Evans 2014: 289). Larvae and adults are usually found underneath coniferous bark.

**Cryptophagidae**

*Caenoscelis ferruginea* (Sahlberg, 1820).—A single specimen of the silken fungus beetle *C. ferruginea* was taken at a UV light at the author’s residence on 22 May 2016. Within the genus *Caenoscelis* Thomson, 1863, as many as twelve species had formerly been described from North America north of Mexico (Leschen 1996), but that number has since been reduced to only four valid species (Leschen and Skelley 2002). *Caenoscelis ferruginea* appears to have a wide and somewhat disjunct (eastern vs. western) distribution, with specimens having been found in Canada: British Columbia; U.S.: Alaska, Indiana, Kentucky, Oregon, Pennsylvania, South Carolina, and Washington (Downey and Arnett 1996b: 1011). Beetles in *Caenoscelis* have the side margins of the pronotum double. In contrast to *C. basalis* Casey, 1900, in which the pronotum is at least one-half wider than long, the pronotum of *C. ferruginea* is narrower (less than one-half wider than long) (Downey and Arnett 1996b: 1010). Both species, however, are pale reddish brown throughout. Like many other members of the family, they feed upon fungus spores in a variety of habitats (e.g., leaf litter, rotting wood).

**Ciidae**

*Octotemnus glabriculus* (Gyllenhal, 1827). – Two specimens of the minute tree fungus beetle *O. glabriculus* were collected in flight at dusk over elm logs at the Oregon Town Park (42° 54’ 7.7”, –89° 25’ 15.6”), Dane County, Wisconsin on dates roughly one year apart, 23 May 2016 and 16 May 2017. These two collecting events both occurred on warm, humid nights. With the synonymy of *O. laevis* Casey, 1898 and *O. glabriculus* (suspected in Thayer and Lawrence 2002: 410; confirmed by Lawrence, pers. comm., 28 May 2017), the genus *Octotemnus* Mellé, 1847 is monospecific. Along with possessing 8 antennomeres, one of its most diagnostic features is the occurrence of spines along the outer edges of more than one-third of the lengths of all tibiae (Thayer and Lawrence 2002: Figs. 19 and 20). Adults and larvae feed upon *Polyporus* fungi, generally under bark and within rotting wood. *O. glabriculus* is widely distributed across North America north of Mexico, occurring within 6 Canadian provinces and 28 U.S. states (Downey and Arnett 1996b: 1117). Its occurrence in Wisconsin is not unexpected.

**Tetrotomidae**

*Eustrophus tomentosus* Say, 1827. – Two specimens of the polypore fungus beetle *Eu. tomentosus* Say, 1827 were collected in similar habitats from two nearby localities. The first specimen was taken from under the bark of a dead-standing elm tree at the author’s residence on 29 April 2012; the second specimen was found under the bark of an elm log at Oregon’s Bicentennial Park (42° 53’ 58.6”, –89° 25’ 15.6”), Dane County, Wisconsin on 7 May 2017. *Eustrophus tomentosus* was formerly classified within *Melandrhyidae* (Downey and Arnett 1996b) but has since been transferred to *Tetrotomidae* (Young and Pollock 2002). The monospecific genus *Eustrophus* Illiger, 1807 is differentiated from the closely-related genus *Eustrophopsis* Champion, 1889 by having finer elytral punctures not forming striae, a slightly lobed posterior margin of the pronotum, and prothoracic episterna lacking a...
transverse suture (Young and Pollock 2002: 414). *Eustrophus tomentosus* has a widespread and somewhat disjunct eastern and western distribution, occurring in Canada: British Columbia; U.S.: Alabama, Indiana, Iowa, Massachusetts, Michigan, New York, North Dakota, Oregon, South Dakota, and Virginia (Downey and Arnett 1996b: 1144). Its occurrence in Wisconsin is not unexpected, either. As their common name implies, these beetles feed upon the fruiting bodies of polycope fungi.

**Curculionidae: Scolytinae**

_Hypothenemus eruditus_ Westwood, 1836. — A single specimen of the bark beetle _H. eruditus_ was collected at a UV light at the author’s residence on 27 May 2018. This species has a worldwide distribution, being found on all continents except Antarctica. _H. eruditus_ represents a species complex, exhibiting a host of cryptic and pseudo-cryptic diversity and has been dubbed the “world’s most common bark beetle” (Kambestad et al. 2017). In North America, it has been reported from 26 continental U.S. states, plus Hawaii, along with Canada and Mexico (Atkinson 2019). In the Midwest, it has been previously known from Michigan, Indiana, Illinois, and Minnesota, and so is not unexpected from Wisconsin. It is absent, however, from the Northwest for reasons not fully understood. _Hypothenemus eruditus_ possesses a vast list of synonyms and more than 300 host records (Atkinson 2019). While a majority of collected specimens are brownish, the Wisconsin specimen is blackish, but possesses the usual rows of stiff flattened interstrial bristles on the elytra and a handful of asperites on the anterior margin of the pronotum. Identification of _H. eruditus_ was provided by Andrew J. Johnson, University of Florida–Gainesville (pers. comm., 29 March 2019).

**Hemiptera**

_Pentatomidae: Asopinae_

**Picromerus bidens** (Linnaeus, 1758). — On 8 September 2018, a single specimen of the introduced predatory stink bug _Pi. bidens_ was captured at a private residence on Fifield Rd., Gillett, Oconto County, Wisconsin. Approximate coordinates of the collecting site are as follows: 44° 57’ 9.66”, –88° 18’35.15”. The specimen had been crawling on a rock. _Picromerus bidens_ possesses a single ventral tooth before the apex of each prothorax. While such femoral teeth are also shared with the genus _Perillus Stål, 1862_, the dark brown dorsal coloration of _Pi. bidens_, with obscure orange spots on the pronotum and base of scutellum, whose tip is pale yellow, should distinguish this genus and species (Swanson et al. 2013).

This Palearctic hemipteran was accidentally introduced to North America from Europe sometime in the early twentieth century, likely on nursery stock or horticultural products (Swanson et al. 2013). While first reported from Vermont (Cooper 1967) and then Canada (Kelton 1972), previously unidentified specimens of the insect were subsequently found to have been collected from Maine in 1932 (Javahery 1986). The species has spread gradually westward, becoming established in other Canadian provinces (as far west as Ontario) and many northeastern U.S. states, including Pennsylvania, by 1999 (Swanson et al. 2013). More recently, _Pi. bidens_ has been found in both the Lower and Upper Peninsulas of Michigan (Swanson et al. 2013, Freese 2014) and Ohio (Chordas 2015). It has an Old World distribution stretching from Ireland to North Africa to Siberia and China (Swanson et al. 2013).

**Lepidoptera**

_Glyphipterigidae_

**Lepidotarphius perornatella** (Walker, 1864). — A single specimen of the newly-introduced sedge moth _L. perornatella_ was captured at a UV light at the author’s residence on 8 August 2018. A photograph of the mounted specimen was posted to BugGuide.net (https://bugguide.net/node/view/1576835/bgimage), where it was identified by Kyhl Austin. The moth’s coloration is striking: the basal three-fifths of the forewings are an emerald green, while the tips are bright orange, containing irregular silver spots. The wingspan is ca. 15 mm. A wide middle band of the abdomen is also orange-colored. The hindwings are a uniform grayish color.

**Lepidotarphius perornatella** was first recognized in North America in August 2017 (Austin et al. 2017). Photographs of live specimens were taken around a small pond in Pella, Marion County, Iowa by Stephen R. Johnson, which were likewise posted to BugGuide.net. Their identity was confirmed from the capture and genitalic dissection of additional specimens by Canadian National Collection entomologists. They belong to the monotypic genus _Lepidotarphius_ Pryer, 1877, although two subspecies, _L. p. perornatella_ and _L. p. fulgens_ (Erschoff, 1877), have been described. _L. perornatella_ is native to China, the Russian Far East, Japan and South Korea, where its larvae feed on the flowering monocot, _Acorus calamus_ Linnaeus (Acoraceae). The genus _Acorus_ is widely used in tropical medicine and horticulture. Importation of this plant was likely the means by
which *L. perornatella* was accidentally introduced to North America (Austin et al. 2017).

**Diptera**

**Phoridae**

*Hirotophora multiseriata* (Aldrich in Brues, 1904).—On 3 July 2018, a single specimen of the native hump-backed fly *Hi. multiseriata* was collected at a UV light at the author’s residence. It was identified by Brian V. Brown, Natural History Museum of Los Angeles County (pers. comm., 10 July 2018). The flattened and enlarged hind femora are elongate-oval with a finely pebbled surface, while the hind tibiae possess a series of transverse rows of hairs along their entire length, for which the species was named. Originally described from Lawrence, Kansas, additional specimens were also recorded from Ithaca, New York (Aldrich in Brues, 1904: 345).¹ Until recently, this species was classified within the subfamily Phorinae and genus *Chaetopleurophora* Schmitz, 1922, where it was one of six known taxa from America north of Mexico (Potenson 1987: 696; Arnett 2000: 883). But a more recent analysis has placed it within the new subfamily Chonocephalinae and new genus *Hirotophora* (Brown et al. 2015). Many phorid larvae are scavengers, whose food preferences range from fungi to carrion; others are parasitic upon various arthropods including social insects. The dietary preference of *Hi. multiseriata* has not been determined.

**Hymenoptera**

**Pteromalidae: Cleonyminae: Ooderini**

*Oodera* sp. – During the course of rearing insects from the cut limbs of a felled shagbark hickory tree, *Carya ovata* (Miller) (Juglandaceae), that originated from the Oregon Town Park, a number of beetle species emerged, along with specimens of multiple families of minute to moderate-sized parasitic wasps. Those included a single specimen of an unidentified species of the introduced pteromalid, *Oodera* sp., that emerged on 20 June 2016. This wasp, ca. 5 mm., possesses enlarged, raptorial profemora that are somewhat analogous to the enlarged, raptorial hind femora of the Chalcididae.

A photograph of the wasp was submitted to BugGuide.net (https://bugguide.net/node/view/1283366/bgimage), where it was identified by Ross Hill as belonging to the Old World genus *Oodera* Westwood, 1874. Additional comments, published by Gary Gibson (2013), accompanied the posting of an earlier specimen of *Oodera* (https://bugguide.net/node/view/737675/bgimage) that had been collected from a Malaise trap on 12 July 2007 by M. J. Hatfield from Story County, Iowa. Gibson noted that *Oodera* was first discovered in the eastern U.S., where it had been reared from the honeylocust *Gleditsia* Linnaeus (Fabaceae) in Maryland, New Jersey, and Virginia. Its known Old World hosts are wood-boring beetles in Buprestidae and Scolytidae. Although it is impossible to positively associate the Wisconsin specimen of *Oodera* sp. with any particular beetle species that emerged from the same hickory limbs, a possible candidate for its host is the small bark beetle, *Chramesus hicoriae* LeConte, 1868 (Curculionidae: Scolytinae), of which several dozen specimens emerged, starting on 17 June. Yet, the purpose of the raptorial profemora of *Oodera* sp. remains unknown.

Apart from the specimen of *Oodera* sp., which was donated as a voucher to the Wisconsin Insect Research Collection (WIRC), all other specimens described herein are retained within the author’s personal collection (JDMC).

**Acknowledgments**

I am indebted to the entomologists at BugGuide.net for providing identifications (either to genera or species) of those insects for which I had submitted photographs. I am also grateful to University of Wisconsin insect diagnostician Patrick J. Liesch for confirming the identities of all insect species named herein with the exception of *Hypothememus eruditus* and *Oodera* sp. Brian V. Brown not only provided the identity but answered further questions regarding the source and date of the original description of *Hirotophora multiseriata* and (presently unknown) food preference. John F. Lawrence (retired) confirmed the synonymy of *Octotemnus laevus* with *O. glabriculus*. Along with furnishing the identity, Andrew J. Johnson provided reference to the paper on *Hypothememus eruditus* by Kambestad et al. (2017).

**Literature Cited**


¹ This publication bears the date 1903 on its cover, but was not released until 1904. B. V. Brown, pers. comm., 16 February 2019.


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New State Record and Range Extension for *Mycterus youngi* Pollock (Coleoptera: Mycteridae)—But Is It Really Rare?

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Abstract

*Mycterus youngi* Pollock (Coleoptera: Mycteridae) was described from Wisconsin and “L.S,” (presumed to indicate along Lake Superior). All but one of the specimens in the type series were collected between 1947 and 1949. Herein, three females of *M. youngi* are reported from Michigan, between 1910 and 1940. A discussion of possible implications of the few, and largely old collection dates is provided.

Keywords: *Mycterus youngi*, Michigan, state record, rarity

Pollock (2012) described *Mycterus youngi* based on seven Wisconsin specimens and two individuals labeled “L.S” only. The latter two specimens, one male and one female, were presumed to have been collected along Lake Superior but had no specific location data and were not provided with collection dates. All but one of the dated specimens in the type series were collected between 1947 and 1949.

While examining undetermined mycterids from the Great Lakes region, two females of *M. youngi* from the Albert J. Cook Collection (Michigan State University: MSUC) and a single female from the Museum of Zoology (University of Michigan: UMMZ) were discovered. These three females are the first Michigan records for this presumably rare species, and by extension the first distributional data points outside Wisconsin (Fig. 1). One specimen was collected from Sanilac County, 4 July [19]28 (MSUC). The second specimen is labeled Atlanta, [Montmorency Co.] MI, 7 July 1940 (MSUC). The third new state record is labeled Mershon Ex.[pedition], Charity Is., Mich. [Arenac County] 17–30 June 1910 (UMMZ).

Ruthven (1911) provided a brief overview of the Mershon Expedition made by the University of Michigan, Museum of Zoology to the Charity Islands and listed A. W. Andrews as the survey member largely responsible for Coleoptera. In addition to Ruthven’s summary, notes and an overview map of the islands in Saginaw Bay, Lake Huron were also provided by Wood (1911) and an ecological overview and preliminary account of the Coleoptera survey were provided by Andrews (1911). Andrews also stated that his actual June visitation dates were 19–26 June (label indicates 17–30 June).

None of the known *M. youngi* specimens aside from a single “flight-intercept trap in sandy barrens” record in the original description (see below) provide any indication of how the specimens were collected and very few plant associations have been provided for any adult *Mycterus* species from North America. Hopping (1935) and Pollock (2002) recorded the western species from flowers of *Ceanothus* (Rhamnaceae) and *Yucca* (Asparagaceae); Pollock (2002) added *Daucus* (Apiaceae), presumably the naturalized and widespread Queen Anne’s lace, *Daucus carota* Linnaeus, on which I have collected *Mycterus scaber* Haldeman in the northern lower peninsula of Michigan. I also have a specimen of *M. scaber* taken on flowers of *Spirea* (Rosaceae) in Quebec, Canada. Adult flight intervals per available collection event label data, excluding the interval associated with the flight intercept trap record (see comments below) and the interval indicated for the “Charity Island” specimen, range from 4 July to 4 August, with mid- to late July being the most common. The collection dates are certainly congruent with the flowering of *D. carota* and several species of *Spirea* in the Great Lakes region.

Even less is known about the microhabitat and food resource requirements of mycterid larvae. Not a single larval association has been confirmed for any North American *Mycterus* species. Larvae presumed to be those of the largely European *Mycterus curculionoides* (Fabricius) were collected beneath bark of *Pinus* sp. (Crowson and Viedma 1964) and this subcortical behavior is consistent with what little is known about larvae of the...
related *Lacconotus* species (Lawrence 1991; personal observations) as well as other eurypine Mycteridae (Pollock 2010).

It is immediately striking that 11 of the 12 known specimens of *M. youngi* came from the 39-year interval between 1910 and 1949. The single outlier is a paratype male recovered from a flight intercept trap 18 years ago (23 June–1 July 2001). These observations beg fundamental questions regarding rarity, vulnerability and extinction risk. Fattorini et al. (2013) provided an overview of “measuring insect rarity” and its contribution to conservation and management. Unfortunately, many of the putatively critical traits they discussed relative to extinction risk (mobility, trophic level, food resource specificity of larvae and adults, other behavioral factors) are virtually unknown for *M. youngi*.

Considering the new Michigan records, the gross geographical range of *M. youngi* would not appear to support a claim of rarity.
From what little is known about resource requirements, it is likely that larvae are saproxylic on coarse woody debris. While such microhabitats would seem to be abundant and not contributing to a case in support of rarity for *M. youngi*, shrinking undisturbed forests and intense forest management provide some cause for concern (Grove 2002, Milberg et al. 2014). Evidence suggests that adults are pollenivorous. *Daucus*, *Spirea* and other mid- to late summer cantharophilous flowers are not at all uncommon in the Great Lakes region, thus precluding adult feeding and potential pre-mating aggregation sites from consideration with respect to possible rarity.

Is *M. youngi* really “rare”? Natural history collections hold the planet’s accumulated historical knowledge relating to biological diversity (Lane 1996, Kemp 2015). Our best regional entomological collections indicate that, if nothing else, *M. youngi* has been rarely collected. But is it rare—is it vulnerable—is it threatened? Perhaps this is merely a case of sampling bias. Then again, there is a plethora of collection records for insects associated with flowers. Pollock (2002) indicated that adults of *Mycterus*, “can be very abundant” on flowers. Perhaps this is another case of being in just the right place at just the right time? An invitation—a challenge.

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